NATIONAL INSTITUTE OF SIDDHA

TAMBARAM SANATORIUM, CHENNAI - 47

THE TAMIL NADU Dr. M.G.R. MEDICAL UNIVERSITY

CHENNAI - 32

Pre-clinical and clinical study on Palagarai Parpam for Hepatoprotective Activity in the management of Kalleral Noi (Liver disease)

&

Pre-clinical and clinical study on Chithiramoola Rasayanam for Anti-Inflammatory Activity in the management of Soolai (Osteoarthritis).

(DISSERTATION SUBJECT)

For the partial fulfillment of the

requirement to the Degree of

DOCTOR OF MEDICINE (SIDDHA)

BRANCH II - GUNAPADAM

APRIL – 2013

BONAFIDE CERTIFICATE

This is certified that I have gone through the dissertation submitted by **Dr.K.Prabhapathy,** (Reg no:32101703) P.G Scholar of **Final Year M.D.(S**), **Department of Gunapadam,** Branch-2, National Institute Of Siddha, Tambaram Sanatorium, Chennai-6000047 and the dissertation work has been carried out by the individual only. This dissertation does not represent or reproduce the dissertations submitted and approved earlier.

Place: Chennai-47

Date:

Prof. Dr.M.Rajasekaran, M.D.(S)

Associate professor,

Head of the Department,

Department of Gunapadam,

National Institute of Siddha,

Chennai-600047.

TRIAL DRUG I PALAGARAI PARPAM

TRIAL DRUG II: CHITHIRAMOOLA RASAYANAM

INTRODUCTION

Kalleral noi (Liver disease) is invariably applied to many disease and disorder, which cause the liver to function improperly and may lead even to stop functioning. Abnormal pain, yellowing of skin or eye (jaundice) or abnormal result of liver function test are the suggestion features of Kalleral noi (Liver disease)[1].

It can caused by many different injuries to liver such as infection with HCV, HBV and other condition like obesity, chronic excessive alcohol consumption, autoimmune disease etc..

Liver disease often lead to lifelong problem and serious consequences. Worldwide liver disease affect hundreds of millions of patients. The increasing incidence of liver disease and liver failure throughout the world have also been reported in India nowadays[2].

At least 1 in 5 Indians is affected with some manifestation of liver diseases. The incidence of liver disease is more in males as compared to females.[3] Statistic reports reveal that nearly 2 lakhs people in India die of liver disease.

Complementary and alternative medicine use is common among patient visiting liver disease clinics in US as in general population (39% Vs 42%) many patients are using to treat their liver disease.[4]

Due to deforestation, urbanisation plants availability are reduced and so we need other than plant product to treat liver disease.

The traditional herbomineral receipe is fast acting powerful hepatic stimulant. It increases the function of capacity of the liver and promotes regeneration and varying positive findings have been reported in several manuscripts. It give support to remarkable improvement in appetite.[5]

Kalleral noi (Liver disease) is due to derangement of Pitham. According to Siddha the bitter taste tends to neutralise the Pitham derangement. Palagarai is a marine drug, which has bitter taste.[6]

In the present scenario, scientific validation and clinical evaluation in tradition medicine system are needed to bridge between tradition medicine system are needed to bridge between tradition and temporary science to reassure the facts said the Ancient.[7]

Hence the author selected Palagarai Parpam which is mentioned in "Kannusami Parambarai Vaithiyam" for Kalleral noi.[8]

AIM:

To evaluate the safety and efficacy of “Palagarai Parpam" for Hepatoprotective activity in the management of Kalleral noi (Liver disease).

OBJECTIVE:

Primary objective:

To evaluate the Hepatoprotective activity of “Palagarai Parpam” for Kalleral noi (Liver disease) in preclinical studies.

Secondary objective:

Biochemical analysis.

To evaluate the efficacy of “Palagarai Parpam” for Hepatoprotective activity in the management of Kalleral noi (Liver disease).

Atomic Absorption Spectrophotometer.

MATERIALS AND METHODS

STANDARD OPERATIVE PROCEDURE:

COLLECTION AND AUTHENTICATION OF RAW DRUGS:

The raw drug was procured from Ramasamy chetty shop Paris Chennai and authenticated by competent authority of department of Gunapadam, National Institute of Siddha.

PREPARATION OF THE MEDICINE:

INGREDIENTS:

Purified Palagarai (Cyperae monetae)

Lemon juice

PURIFICATION METHODS:[9]

Purification of Palagarai:

Palagarai soaked in lemon juice and washed well in water.

METHOD OF MEDICINE PREPARATION:[8]

Palagarai soaked in lemon juice for one night and then covered with mud cap with clay pasted cloth. Then it was calcinated with 30 – 40 cow dung cake. Uncover the mud cap and grind the outcome as fine powder. The powder was stored in a clean ,dry air tight glass bottle.

LABELLING:

Name of the preparation	: Palagarai Parpam
Quantity of the drug	: 2gm
Dose	:130 mg, twice a day
Adjuvant or Vehicle	: Ghee
Indication	: Palagarai Parpam
Date of manufacturing	:1/4/12
Date of expiry	:100 years from the date of manufacture[9]

Palagarai (Cypraea moneta)

Palagarai Parpam

REVIEW OF THE LITERATURE

GUNAPADAM ASPECT

பலகறை [10]

வேறுபெயர் : கவடி, சோகி, வராடி

"மண்ணிய கவடி சோகி வராடியே பலகறைப்பேர்"

சுவை : கைப்பு

செய்கை : தாதுவெப்பகற்றி, கோழையகற்றி, வெப்பகற்றி

பொதுகுணம் :

"மந்தந்தா கங்கிரகணி மாவிடச் சுரங்கண்ணோய்
தொந்தம் பரிநாமச் சூலைகய – மிந்த
வுலகறையைக் காலோடிவை யோடுநரைத்த
பலகறையை காணினியம் பார்"

குணம் :

அலசம், தாகம், கிரகணி, மகாவிட சுரங்கள், விழிநோய், வாத தொந்தம், பலவிதக் குத்தல் கயம், கபவாதம், அஜீரணம், காமாலை, கல்லீரல், மண்ணீரல், வீக்கம், சுவாசகாசம், காசம் முதலிய நோய்கள் தீரும்.

சுத்தி முறை :[9]

1. ஒரு பலம் பலகறை பொடிக்கு பத்துபலம் தமரத்தம் பழச்சாறு காலையில் விட்டு மாலை வரை வெயிலில் வைத்து எடுத்து மறுநாள் காலையிலும் புதிதாக மேற்படி சாற்றை விட்டு வெய்யிலில் வைக்கவும். இதுபோல் பதினைந்து நாட்கள் செய்தெடுக்க சுத்தியாகும்.
2. பலகறையை எலுமிச்சை பழச்சாற்றிலேனும் அரிசிக் கஞ்சியிலேனும் ஊறவைத்தெடுக்க சுத்தியாகும்.

எலுமிச்சை[10]

(Citrus acidica)

சுவை : புளிப்பு, கார்ப்பு, **தன்மை :** வெப்பம், **பிரிவு :** கார்ப்பு

செய்கை : குளிர்ச்சியுண்டாக்கி

குணம் :

"தாகம் குநகநோய் தாழாச் சிலிபதநோய்
வேகங்கொள் உன்மாதம் வீறுபித்தம் - மாகண்ணோய்
கன்னனோய் வாந்தியும்போங் கட்டுவா தித்தொழிலில்
மன்னெலுமிச் சங்கனியை வாழ்த்து"

இது, மயக்கம், வாந்தி, வாய்க்குமட்டல், நீர்வேட்கை, வெறி, கண்ணோய், காதுவலி இவைகளைப் போக்கும்; நகச்சுற்றுக்கும் நன்மை தரும்.

சேரும் மருந்துகள்

பலகறை பற்பம்:

அளவு :1-1 1/2 குன்றி

அனுபானம் :எலுமிச்சை பழச்சாறு

தீரும் நோய் : ஈரல்களின் வீக்கம்

ஆதாரம் :பதார்த்த குணவிளக்கம்

பலகறை பற்பம்:

அளவு :1-1 1/2 குன்றி

அனுபானம் :வெண்ணெய்/நெய்

தீரும் நோய் : வயிற்று நோய், மகோதரம், உப்புசம், பெருவயிறு

ஆதாரம் : அனுபோக வைத்திய நவநீதம்

பலகறை பற்பம்:

அளவு :1-2 குன்றி

அனுபானம் : வெண்ணெய்/நெய் /தேன்

தீரும் நோய் : பித்தசமந்தமான நோய்

ஆதாரம் :அனுபோக வைத்திய நவநீதம்

பலகறை பற்பம்:

அளவு :2-4 குன்றி

அனுபானம் : வெண்ணெய்/நெய் /தேன்

தீரும் நோய் : வயிற்று வலி

ஆதாரம் :அனுபோக வைத்திய நவநீதம்

ZOOLOGICAL ASPECT

Cypraea moneta[11]

Cypraea moneta (money cowry), is a species of a marine gastropod mollusk in the family Cypraeidae, . This cowry lives in intertidal rocky areas and shallow tide pools among sea weed, coral remains, and empty bivalve shells. It can be found on and under rocks in shallow water and on exposed reefs at low tide. It feeds on algae and marine vegetation growing on loose rocks and pieces of dead coral.

ZOOLOGICAL NAME: Cypraea moneta

Classification[12]

Kingdom : Animalia
Phylum :Mollusca
Class :Gastropoda
Orderd : Sorbeoconch
Superfamily : Cypraeoidea
Family :Cypraeidea
Genus : Cypraea
Species : Cypraea moneta

VERNACULAR NAME:[13]

ENGLISH : Cowry
SANSKRIT : Varatika
HINDI : Cowries
TELUNGU : Gavala
MALAYALAM : Kavadi

DESCRIPTION:

The shell is 30 to 45 mm long. It is white to straw-colored.[1] Small, convoluted ,glossy shell of variegated colours, oblong oval shape varying in size from a tamarind seed to an almond. The upper surface is smooth, shining and convex. Base is compressed with a cleft in the centre which is runs longitudinally. The margin of the cleft is serrated on one side and other side is depressed.

DISTRIBUTION:

This is a very common species which is found widely in Indo-Pacific tropical waters. It is present in numerous regions, including East and South Africa, Madagascar, the Red Sea and the Persian Gulf, eastern Polynesia, Galapagos, Clipperton and Cocos islands off Central America, southern Japan, Midway and Hawaii, and northern New South Wales and Lord Howe Island.

VARITIES:

White, red, yellow. Different colours of Cowry are dependent on the sexual hormone, genetic factors, pigmentation, disease, injury, diet, Presence or absence of aluminium and other compounds , the acidity of the soil and water[15].

Ancient Hindu alchemists preferred shells which were yellow colour, knotty and possessed of circular lines on dorsal side[13].

PHYSICAL CONSTANTS:

Insoluble in water, soluble in hydrochloric acid with effervescence.

CHEMICAL CONSTITUENTS:

Phosphate, flouride and carbonate of calcium, magnesium phosphate, mangenese and sodium chloride.

PHARMACOLOGICAL ACTIVITIES:[16]

Anti- spasmodic activity, anti -microbial activity.

எலுமிச்சை

BOTANICAL NAME : Citrus acida[17]

VERNACULAR NAME :

ENGLISH : Lime

SANSKRIT : Jambira

HINDI : Nimbu limu

TELUNGU : Nimma

MALAYALAM : Cheru-naranga

BOTANICAL DESCRIPTION :

Fruits usually small, globose or ovoid ;rind thick or thin; pulp pale, very acid.

PARTS USED:

Fruit

PHARMACOLOGICAL ACTIVITIES:[18]

USES;

The fruit has a sour sharp taste ;appetiser, stomachic ,anthelmintic, cures abdominal complains, removes diseases due to "thridosha", loss of appetite, constipation, fatigue, good in" kapha "and biliousness, abdominal pain, and foul breath; relieves vomiting; good for the eyes.

The fruit has a sour sharp taste; with flavours; stimulant: useful in weakness and trembling of the limbs hemicrania, throat troubles, brain disorder , relieves biliousness, vomiting, retching, improves liver, heart.

Supportive Journals:

The analysis of the Cowry bhaspam shows that the overall decarbonation of calcium carbonate in argonite form and reformation of calcium carbonate in the calcinate form. The transformation occur via formation of Calcium hydroxide and Calcium oxide as intermediate. Cowry bhaspam is highly crystalline Calcium carbonate ih the calcite form with presence of trace element like Mg, Al, K, Fe. It Pharmacological activity shows that 10% of all Cowries had been investigated in detail for bioactive agent.[19]

Cowry Bhaspam contain Phosphate, Fluoride and Carbonate of Calcium , Magnesium Phosphate and Manganese.[20]

PHYSICAL PROPERTIES

Materials and Methods

The Physical properties of Palagarai Parpam were analysed in the following procedure at Sri Ramachandra University.

pH at 10% of aqueous solution:

Five grams of Palagarai Parpam was weighed accurately and placed in clear 100 ml beaker. Then 50 ml of distilled water was added to it and dissolved well. After 30 minutes it was then applied in to pH meter at standard buffer solution of 4.0,7.0,9.2. (Trail Drug 1Table -2).

Ash Values:

The Ash values measures-inorganic constituents present in the raw drug. A high ash content explains its unsuitable nature to be used as a drug (Trail Drug1 Table -2).

Total Ash

A little of extract was taken in a silica crucible previously ignited, cooled and weighed. It was incinerated by gradually increasing the heat not exceeding dull red heat (450°C) until free from carbon, cooled and weighed. The percentage of ash was calculated with reference to air- dried drug. The procedure was repeated to get the constant weight.

(TRAIL Drug 1 Table -2)

Water soluble ash

The total ash was boiled with 25 ml water and filtered through ash less filter paper (Whatmann 4.1). It was followed by washing with hot water .The filter paper was dried and ignited in the silica crucible, cooled and the water insoluble ash was weighed. The water-soluble ash can be calculated by subtracting the water insoluble ash from the total ash.

Acid insoluble ash

The total ash obtained was boiled for 5 minutes with 25 ml of (10% w/v) dilute hydrochloric acid and filtering through ash less filter paper (Whatmann 4.1). The filter paper was ignited in the silica crucible, cooled and insoluble ash was weighed.*(* Trail Drug 1 Table -2)

BIO -CHEMICAL ANALYSIS OF PALAGARAI PARPAM

The biochemical analysis of the **Palagarai Parpam** was carried out in the Biochemistry lab, NIS

. S.No	EXPERIMENT	OBSERVATION	INFERENCE
1.	Appearance of sample	Light white in colour	
2.	**Solubility:** a. A little(500mg) of the sample was shaken well with distilled water. b. A little(500mg) of the sample was shaken well with con. HCl/Con. H_2So_4	Sparingly soluble	Absence of Silicate
3.	**Action of Heat:** A small amount(500mg) of the sample was taken in a dry test tube and heated gartly at first and then strong.	white fumes evolved	Presence of Carbonate
4.	**Flame Test:** A small amount(500mg) of the sample was made into a paste with con. HCl in a watch glass and introduced into non-luminous part of the Bunsen flame.	No Bluish green flame appeared.	Absence of Copper
5.	**Ash Test:** A filter paper was soaked into a mixture of sample and dil. cobalt nitrate solution and introduced into the Bunsen flame and ignited.	Yellow colour flame appeared.	Presence of sodium

Preparation of Extract:

5gm of Palagarai Parpam was weighed accurately and placed in a 250ml clean beaker and added with 50ml of distilled water. Then it was boiled well for about 10 minutes. Then it was cooled and filtered in a 100ml volumetric flask and made up to 100ml with distilled water.

S.No	EXPERIMENT	OBSERVATION	INFERENCE
	I.Test For Acid Radicals		
1.	**Test For Sulphate:** a. 2ml of the above prepared extract was taken in a test tube to this added 2ml of 4% dil ammonium oxalate solution b. 2ml of the above prepared extracts was added with 2ml of dil-HCl was added until the effervescence ceases off. Then 2ml of dil.Barium chloride solution was added.	Cloudy appearance present	Absence of Sulphate
2.	**Test For Chloride:** 2ml of the above prepared extract was added with dil. HCl till the effervescence ceases. Then 2ml of dil.silver nitrate solution was added.	Cloudy appearance presence.	Presence of Chloride
3.	**Test For Phosphate:** 2ml of the extract was treated with 2ml of dil.ammonium molybdate solution and 2ml of con.HNo3.	No yellow appearance present	Absence of Phosphate
4.	**Test For Carbonate:** 2ml of the extract was treated with 2ml dil. Magnesium sulphate solution	Cloudy appearance presence.	Presence. of carbonate
5.	**Test For Nitrate:** 1gm of the substance was heated with copper turning and concentrated H2So4 and viewed the test tube vertically down.	No Brown gas evolved.	Absence of Nitrate

6.	**Test For Sulphide:** 1gm of the substance was treated with 2ml of con. HCL	No Rotten Egg Smelling gas.	Absence of Sulphide
7.	**Test For Fluoride & Oxalate:** 2ml of extract was added with 2ml of dil. Acetic acid and 2ml dil.calcium chloride solution and heated.	No Cloudy appearance	Absence of fluoride and oxalate
8.	**Test For Nitrite:** 3drops of the extract was placed on a filter paper, on that-2 drops of dil.acetic acid and 2 drops of dil.Benzidine solution was placed.	No Characteristic changes	Absence of Nitrite
9.	**Test For Borate:** 2 Pinches(50mg) of the substance was made into paste by using dil.sulphuric acid and alcohol (95%) and introduced into the blue flame.	No Bluish green colour flame.	Absence of borate
	II. Test For Basic Radicals		
1.	**Test For Lead:** 2ml of the extract was added with 2ml of dil.potassium iodine solution.	No yellow Precipitate obtained.	Absence of Lead
2.	**Test For Copper:** One pinch(50mg) of substance was made into paste with con. HClin a watch glass and introduced into the non-luminuous part of the flame.	No Blue colour flame. No Blue colour precipitate formed.	Absence of copper
3.	**Test For Aluminium:** To the 2ml of extract, dil.sodium hydroxide was added in 5 drops to excess.	No Yellow colour appeared.	Absence of aluminium

4.	**Test For Iron:** a. To the 2ml of extract,2ml of dil.ammonium solution was added. b. To the 2ml of extract 2ml thiocyanate solution and 2ml of con HNo3 was added	blood red colour appeared.	Presence of Iron
5.	**Test For Zinc:** To 2ml of the extract, dil.sodium hydroxide solution was added in 5 drops to excess and dil.ammonium chloride was added.	No White precipitate was formed	Absence of Zinc
6.	**Test For Calcium:** 2ml of the extract was added with 2ml of 4% dil.ammonium oxalate solution	Cloudy appearance and white precipitate was obtained	Presence. of calcium
7.	**Test For Magnesium:** To 2ml of extract dil.sodium hydroxide solution was added in drops to excess.	White precipitate was obtained	Presence of Magnesium
8.	**Test For Ammonium:** To 2ml of extract 1 ml of Nessler's reagent and excess of dil.sodium hydroxide solution were added.	No Brown colour appeared	Absence of ammonium
9.	**Test For Potassium:** **A pinch(25mg) of substance was treated of with 2ml of dil.sodium nitrite solution and then treated with 2ml of dil.cobalt nitrate in 30% dil.glacial acetic acid.**	No Yellowish precipitate was obtained.	Absence of Potassium
10.	**Test For Sodium:** **2 pinches(50mg) of the substance was made into paste by using HCl and introduced into the blue flame of Bunsen burner.**	yellow colour flame appeared	Presence of sodium

11.	**Test For Mercury:** **2ml of the extract was treated with 2ml of dil.sodium hydroxide solution.**	No yellow precipitate was obtained	Absence of mercury
12.	**Test For Arsenic:** **2ml of the extract was treated with 2ml of dil.sodium hydroxide solution.**	No brownwish red precipitate was obtained	Absence of arsenic
	III. Miscellaneous		
1.	**Test For Starch:** **2ml of extract was treated with weak dil.iodine solution**	No blue colour developed	Absence of starch
2.	**Test For Reducing Sugar:** **5ml of Benedict's qualitative solution was taken in a test tube and allowed to boil for 2 minutes and added 8 to 10 drops of the extract and again boil it for 2 minutes. The colour changes are noted.**	Brick red colour not developed	Absence of reducing sugar
3.	**Test For The Alkaloids:** **a) 2ml of the extract was treated with 2ml of dil.potassium iodide solution.** **b) 2ml of the extract was treated with 2ml of dil.picric acid.** **c) 2ml of the extract was treated with 2ml of dil.phosphotungstic acid.**	Yellow colour developed	Presence of Alkaloid
4.	**Test For Tannic Acid:** **2ml of extract was treated with 2ml of dil.ferric chloride solution**	black precipitate was obtained	Absence of Tannic acid
5.	**Test For Unsaturated Compound:** **To the 2ml of extract 2ml of dil.Potassium permanganate solution was added.**	Potassium permanganate was not decolourised	Absence of unsaturated compound

6.	**Test For Amino Acid:** **2 drops of the extract was placed on a filter paper and dried well. 20ml of Biurette reagent was added.**	violet colour developed	Presence of amino acids
7.	**Test For Type Of Compound:** **2ml of the extract was treated with 2 ml of dil.ferric chloride solution.**	No green colour developed No red colour developed No violet colour developed No blue colour developed	Absence of oxy quinole pinephrine and pyro catechol Anti pyrine, Aliphatic amino acids and meconic acid are absent Apomorphine salicylate and Resorcinol are absent Morphine, Phenol cresol and hydro uinone are absent

Elemental Analysis using Atomic Absorption Spectrophotometer.

(TRAIL DRUG 1 ,TABLE 3)

The AAS of Palagarai Parpam was done at Sri Ramachanra University. In this method the sample, in the form of a homogeneous liquid, is introduced into a flame where thermal and chemical reactions create "free" atoms capable of absorbing, emitting or fluorescing at characteristic wavelengths.

In Atomic Absorption Spectrophotometer (AAS) the majority of free atoms in the commonly used flames were in the ground state, but that the flames did not also have enough energy to excite these atoms. A light source emitting a narrow spectral line of the characteristic energy is used to excite the free atoms formed in the flame. The decrease in energy (absorption) is then measured.

METHODOLOGY

I. Microwave Digestion For Elemental Analysis

Model Name: Multiwave3000

Digestion Procedure:

I. 200mg of the given sample is placed in a digestion vessel, acid is added and the mixture is heated for several minutes. After the digestion, the samples are diluted to a specific volume. If too much sample is used in wet digestion, the reaction mixture can become violent. The samples are placed in digestion vessels that fit directly into digestion racks. There are several different acids or mixtures of acids used for digestion, the most common of which is concentrated Hydrochloric acid. The samples are heated slowly at a high temperature. After digestion, the samples are diluted to the appropriate volume with deionized H_2O.

II. Elemental Analysis using Atomic Absorption Spectrophotometer

The elemental analysis of digested samples have been determined by Atomic Absorption Spectrophotometer- Flame technique (AAS model 400 Perkin Elmer). Working standard solutions of Fe, Mg and Zn were prepared from stock standard solution of 1000 ppm from MERCK. Using blank solution to zero the instrument performs the Calibration. The standards are then analyzed and their absorbance recorded. A graph of Absorbance Vs

Concentration is plotted. Calibration of the instrument was repeated periodically during operation. A blank reading was also taken and necessary correction was made during the calculation of concentration of various elements.

The digested material was made upto 100 ml for analysis in an (AAS) atomic absorption spectrophotometer (Perkin Elmer). The results were calibrated using standard calibration curve.

In AAS the Wave Length (nm), Flame type, Lamp source and Calibration range (ppm) of different elements have been used, are listed in table.

Instrumental conditions for elemental analysis

Element	Wavelength nm	Light source	Flame type
Magnesium	**285.2**	**HCL**	**Air/Ac**
Iron	**386.0**	**HCL**	**Air/Ac**
Zinc	**213.86**	**HCL**	**Air/Ac**

Air/Ac: Air-Acetylene; HCL: Hallow cathode lamp

Elemental Analysis using Flame photometer

The analysis of Na, K and Ca of the digested samples have been determined by Flame photometer (Flame photometer 129- Systronics Make). see trail drug 1 table 3

TOXICITY STUDY

ACUTE AND SUB ACUTE TOXICITY STUDY ON PALAGARAI PARPAM IN RODENTS.

Animals

Mice of either sex weighing 25-30g and rats weighing 210-240g were obtained from the animal house of Vels University. The animals were used with the approval of the Institute animal ethics committee and obtained from Vels University, Chennai. They were fed with a balanced standard pellet diet and maintained under standard laboratory conditions, providing 24-28^{0}C temperature, standard light cycle (12 h light, 12 h dark) and water ad libitum.

Animals were kept in cages with raised floors of wide mesh to prevent coprophagy. Animal welfare guidelines were observed during the maintenance period and experimentation. The rats were randomly assigned to control and different treatment groups, six animals per group. The animals were acclimatized for one week under laboratory conditions.

ACUTE TOXICITY STUDY-OECD 425 GUIDELINES

Acute oral toxicity test for the Palagarai Parpam was carried out as per OECD Guidelines 425. As with other sequential test designs, care was taken to ensure that animals are available in the appropriate size and age range for the entire study. The test substance is administered in a single dose by gavage using a stomach tube or a suitable intubation cannula. The fasted body weight of each animal was determined and the dose was calculated according to the body weight. After the substance has been administered, food was withheld for a further 2 hours in mice. The animals were observed continuously for the first 4 h and then each hour for the next 24 h and at 6 hourly intervals for the following 48 h after administering of the test drug, to observe any death or changes in general behaviour and other physiological activities.

Single animals were dosed in sequence usually at 48 h intervals. However, the time interval between dosing is determined by the onset, duration, and severity of toxic signs. Treatment of an animal at the next dose was delayed until one is confident of survival of the previously dosed animal.

Observation of toxicity signs: General behaviour, respiratory pattern, cardiovascular signs, motor activities, reflexes, change in skin and fur, mortality and the body weight changes were monitored daily. The time of onset, intensity, and duration of these signs, if any, was recorded.

Acute Toxicity study

Acute toxicity study was performed for Palagarai Parpam according to the acute toxic up and down method as per OECD guidelines 425, albino mice were used for acute toxicity study. The animals were kept fasting for overnight providing only water, after which the Palagarai Parpam was administered orally at the dose of 250, 500, 1000, 2000 mg/kg and observed for 14 days. If mortality was observed in animals, then the dose administered was assigned as toxic dose. If the mortality was observed in animal, then the same dose was repeated again to confirm the toxic dose (TRIAL DRUG1 TABLE 5)

SUB-ACUTE TOXICITY

In a 28-days sub acute toxicity study, twenty four either sex rats were divided into four groups of 6 rats each. Group I that served as normal control was administered with distilled water (p.o.) while groups II, III and IV were administered daily with the Palagarai Parpam (p.o.) for 28 days at a dose of 25mg, 50mg, 100mg/kg respectively. The animals were then observed daily for gross behavioural changes and any other signs of subacute toxicity.

The weight of each rat was recorded on day 0 and weekly throughout the course of the study, food and water consumption per rat was calculated. At the end of the 28 days they were fasted overnight, each animal was anaesthetized with diethylether, following which they were then dissected and blood samples were obtained by cardiac puncture into heparinised tubes. The blood sample collected from each rat was centrifuged with 3000 X g at $4^{o}C$ for 10 min to separate the serum and used for the biochemical assays.

Hematological and blood biochemical analysis:

At the end of the study, all animals were kept fasted for 16-18 h and then anesthetized with anesthetic ether on the 28th day. Blood samples for hematological and blood chemical analyses were taken from retro orbital vein. Heparinized blood samples were taken for determining complete blood count (white blood cell count, differential white blood cell count, platelet count, red blood cell count, hematocrit, and hemoglobin) by semiautomated hematology analyzer.

The serum from non-heparinized blood was carefully collected for blood chemistry and enzyme analysis like glucose, Creatinine, Total protein, Albumin, Total and Direct bilirubins, Serum glutamate-oxaloacetate transaminase (SGOT), Serum glutamate pyruvate transaminase (SGPT), and Alkaline phosphatase (ALP) were automatically determined using autoanalyzer.

Necropsy:

All rats were sacrificed after the blood collection. The positions, shapes, sizes and colors of internal organs were evaluated. The Spleen, Testes, Pancreas, Lung, Liver, Brain, Heart, Stomach, Intestine, Bone, Ovary, and Kidney tissues were excised from all rats to visually detect gross lesions, and weighed to determine relative organs' weights and preserved in 10% neutral formalin for histopathological assessment. The tissues were embedded in paraffin, and then sectioned, stained with haematoxylin and eosin and were examined microscopically.

Statistical analysis (Trial Drug 1 Table 6-13)

Values were represented as mean ± SEM. Data were analysed using one-way analysis of variance (ANOVA) and group means were compared using the Tukey-Kramer Multiple Comparison test using Instat-V3 software. P values < 0.05 were considered significant.

RESULTS AND DISCUSSION

Animals were shown negligebale toxic Signs during the dosing period of 28 days. Results of body weight determination of animals of control and different dose groups exhibited comparable body weight gain throughout the dosing period of 28 days. During dosing period, the quantity of food consumed by animals from different dose groups was found to be normal with that of control animals. Ophthalmoscopic examination of animals in control and test product– treated groups did not reveal any major and remarkable abnormality.

Urine analysis data of control group and treated group of animals determined in week 4 and animals in week 6 did not reveal any remarkable abnormalities except few indications. Comparison of organ weights of treated animals with respective control animals on day 28 was found to be comparable except brain and kidneys. Gross pathological examination of animals in control as well as the treated groups revealed mild abnormalities like kidney damage.

The results of haematological investigations conducted on day 28, revealed no significant changes in the values of different parameters investigated when compared with those of respective controls; However, the increase or decrease in the values obtained was within normal biological and laboratory limits. A slight increase in total RBC count values were obtained for animals in the dose group of 50 and 100 mg/kg and also decreased values of platelets ($P>0.05$) were observed for animals in dose groups administered 100mg/ kg body weight of Palagarai Parpam sacrificed on day 28.

The results of Biochemical investigations conducted on days 28 revealed the significant changes in the values of ALP, Globulin, Urea, creatinine and uric acid at 50 and 100 mg/kg dose level particularly when compared with those of respective controls. HDL, LDL, Triglyceride levels are elevated in animals of 25 - 100 mg/kg dose group ($P<0.01$). Glucose level decreased in animals of the entire drug treated group ($P<0.01$).

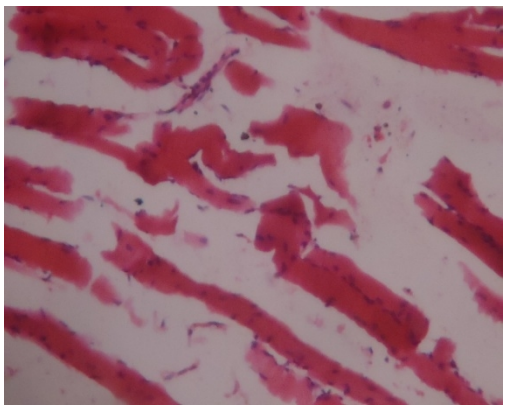
CONCLUSION:

The results indicated the toxic effect at 500mg/kg of Palagarai Parpam *treated* via oral route over a period of 28 days. So, it can be concluded that the Palagarai Parpam can be prescribed for long term therapeutic use in human with the necessary dosage reduction and can be used up to 250mg/kg. body weight p.o.

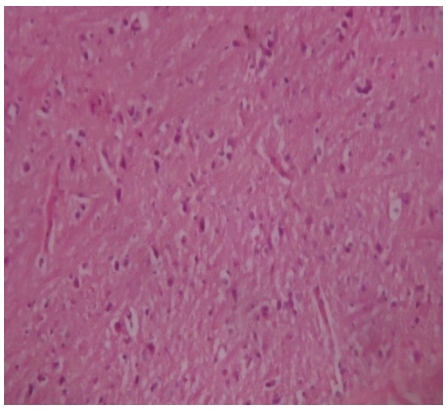
PALAGARAI (Cypraea moneta)

HISTOPATHOLOGY

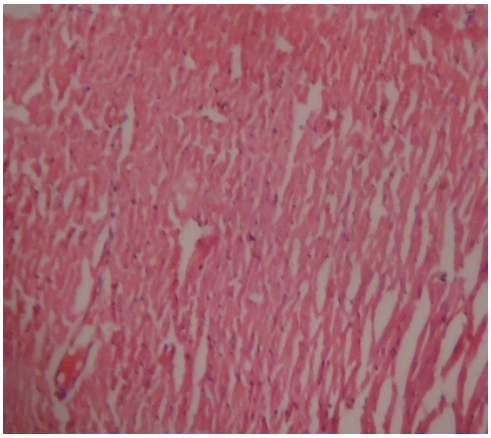
Bone



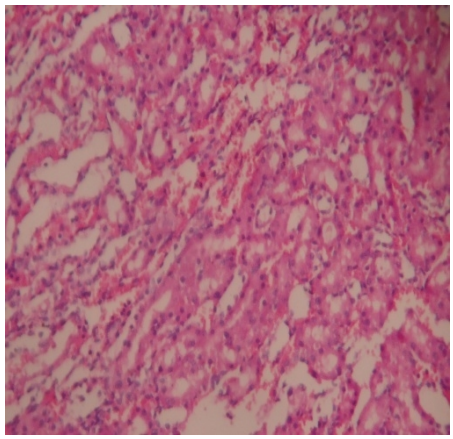
Brain



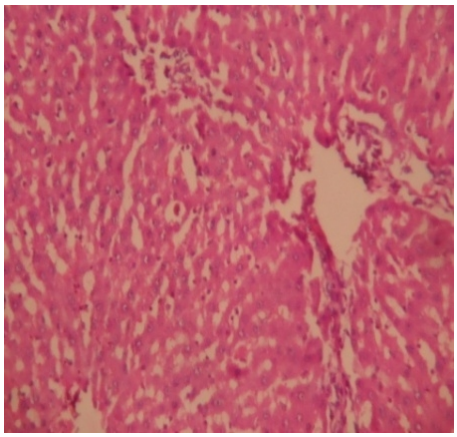
Heart



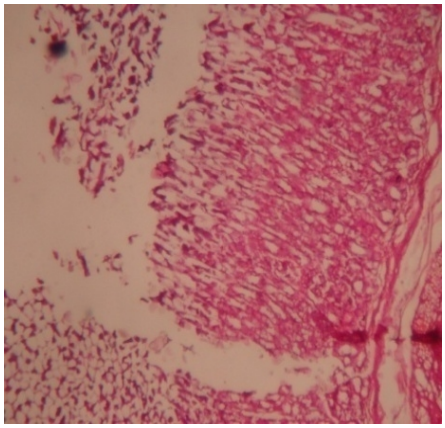
Kidney



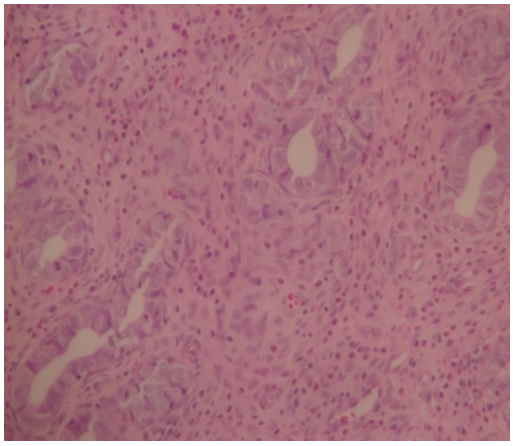
Liver



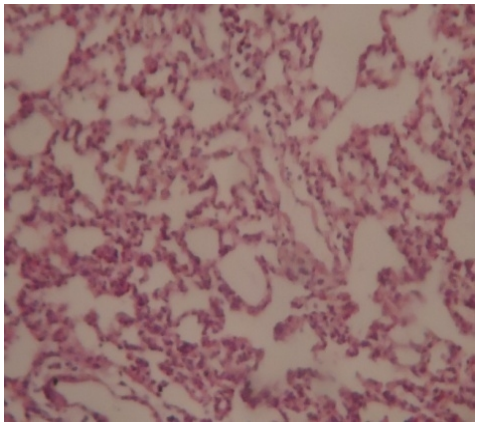
Intestine



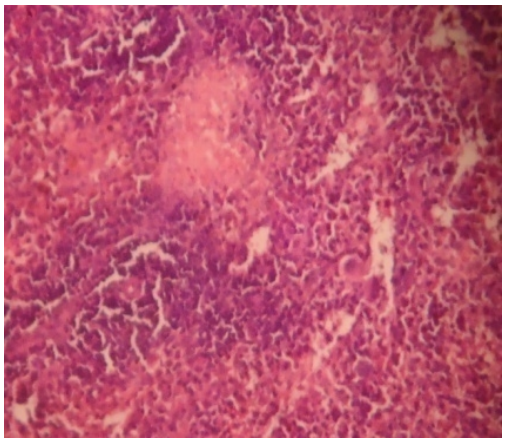
Ovary



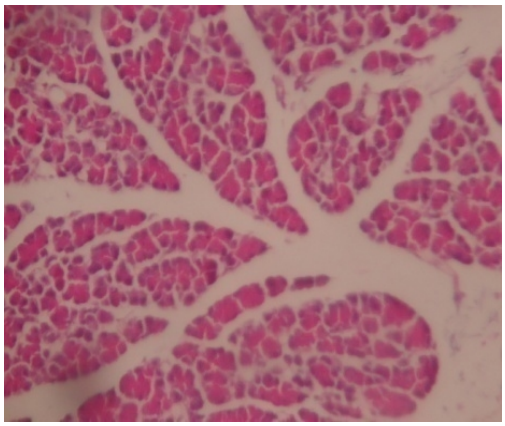
Lung



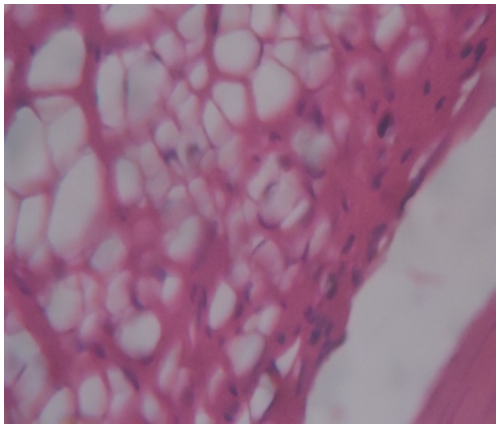
Spleen



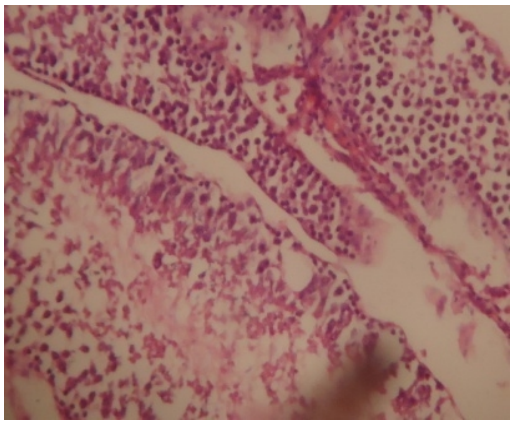
Pancreas



Stomach



Testis



PHARMACOLOGICAL STUDY

HEPATOPROTECTIVE ACTIVITY OF PALAGARAI PARPAM AGAINST CCL4 INDUCED HEPATOTOXICITY RATS

INTRODUCTION

Liver diseases remain one of the most serious health problems. In view of severely undesirable side effects of synthetic agents and the absence of reliable liver-protective drugs in modern medicine, there are a number of medicinal preparations in the Siddha system of Indian medicine recommended for the treatment of liver disorders. Their usage has been popular for centuries and are quite often claimed to offer significant relief. Also, there is a growing trend to follow systematic research methodology and to evaluate the scientific basis for traditional medicines that are claimed to possess hepatoprotective potential.

As the efficacy of Siddha medicinal products in preclinical liver diseases is not well documented, accurate scientific assessment has become a prerequisite for acceptance of health claims. In the present study, an attempt has been made to study the effect of Palagarai Parpam in rats. Liver is the key organ regulating homeostasis in the body. It is involved with almost all the biochemical pathways related to growth, fight against disease, nutrient supply, energy production and reproduction. Because of its unique metabolism and relationship to the gastrointestinal tract, the liver is an important target for toxicity produced by drugs, xenobiotics and oxidative stress. More than 900 drugs, toxins and herbs have been reported to cause liver injury and drugs account for 20-40% of all instances of fulminant liver failure.

In the absence of reliable liver protection drugs in modern medicine, a large number of medicinal preparations are recommended for the treatment of liver disorders and quite often claimed to offer significant relief. Attempts are being made globally to get scientific evidences for these traditionally reported Siddha drugs. This scenario provides a severe necessity to carry out research in the area of hepatotoxicity.

MATERIALS AND METHODS

Drugs and chemicals

Silymarin was a gift sample from Micro Laboratories, Hosur, India. Aspartate amino transferase (ASAT) and alanine amino transferase (ALAT), alkaline phosphatase (ALP) and total proteins (TP) kits were from RANDOX Laboratories Ltd. All other chemicals and reagents used were of analytical grade.

Animals

Male Wistar rats weighing between 150 – 220 gm were used for this study. The animals were obtained from animal house, Department of Pharmacology, School of Pharmaceutical Sciences, Vels University, Tamilnadu, India. The animals were placed at random and allocated to treatment groups in polypropylene cages with paddy husk as bedding. Animals were housed at a temperature of 24±2^{o}C and relative humidity of 30 – 70 %. A 12:12 light: day cycle was followed. All animals were allowed to free access to water and fed with standard commercial pelleted rat chaw (M/s. Sai durga foods, Bangalore). All the experimental procedures and protocols used in this study were reviewed by the Institutional Animal Ethics Committee and were in accordance with the guidelines of the IAEC. (Approval number: XIII/VELS /PCOL/33/2000/C PCSEA/IAEC / 08.08.2012).

Hepatoprotective Activity:

A total of 30 animals were equally divided into 5 groups of six each.

Group I: served as normal control received 0.5% (CMC) carboxy methyl cellulose solution (1 ml/kg) once daily (untreated);

Group II: served as CCL4 control, administered with Carbon tetrachloride 3 ml / Kg.

Group III: CCl4 + Palagarai Parpam (50mg/kg) treated animals;

Group IV: CCl4 + Palagarai Parpam (100mg/kg) treated animals.

Group V: served as reference control, received CCl4 + Silymarin (100mg/kg) once daily.

All the test drugs and Standard were administered orally by suspending in 0.5% CMC solution. After 48h of CCL4 administration and after the last dose of the test drugs, the blood was collected by retro orbital puncture under light ether anesthesia and serum was separated for the estimations of alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP) and bilirubin.

Histopathological evaluation

At the end of the study, all the surviving animals of the respective groups were sacrificed by an overdose of ether anaesthesia. After exsanguinations of the animal's liver were removed immediately and washed with ice-cool saline. The tissue samples were fixed with 10% formaldehyde, dehydrated in a graded series of ethanol, and embedded in paraffin wax before sectioning. The tissue was cut into sections approximately 5 μm thick, dewaxed, and rehydrated. The sections were then stained with haematoxylin-eosin dye and studied for histopathological changes using a light microscope. Each sample was observed at a magnification of 100X.

Statistical Analysis: (Trial Drug 1 Chart 1-3)

The values were expressed as mean ± SEM. The statistical analysis was carried out by one way analysis of variance (ANOVA) followed by Dunnet's 't' - test. P values <0.05 were considered significant.

RESULTS AND DISCUSSION

Carbon tetrachloride is a routinely used hepatotoxin for experimental study of liver diseases. Administration of CCl_4 causes acute liver damage that mimics natural causes. It mediates changes in liver function that ultimately leads to destruction of hepatocellular membrane. Cytochrome P-450 activates CCl_4 to form various free radicals (trichloromethyl, Cl_3 C-CCl_3 (hexachloroethane), $COCl_2$ (phosgene), etc.) which are involved in the pathogenesis of liver damage in chain reactions resulting in peroxidation of lipids, covalent binding of macromolecules, disruption of metabolic mechanisms in mitochondria, decreasing levels of phospholipids, increasing triglyceride levels, inhibition of calcium pumps of microsomes thus leading to liver necrosis. Liver plays a major role in detoxification and excretion of many endogenous and exogenous compounds, any injury or impairment of its function may lead to several implications on one's health. Management of liver diseases is still a challenge to modern medicine.

Conventional drugs used in the treatment of liver diseases are often inadequate. It is therefore necessary to search for alternative drugs for the treatment of liver diseases to replace the currently used drugs of doubtful efficacy and safety. The hepatotoxic effect of CCl4 has been reported to be due to its metabolite CCl3º, a free radical that alkylates cellular proteins and other macromolecules. This study shows that hepatic injury induced by CCl4 caused significant rise in marker enzymes SGOT, SGPT, ALP and total bilirubin

.The serum enzymes like SGOT, SGPT, ALP and total bilirubin of treated animals were significantly reduced ($p<0.01$) by treatment of Palagarai Parpam at two dose levels 50mg/kg and 100mg/kg , when compared with CCl4 treated control. From the result it is clear that the drugs show dose dependent activity. In histological studies, hepatocytes of the normal control group showed a normal lobular architecture of the liver. Whereas, the CCL4 treated group the liver showed hepatocytic necrosis and inflammation also observed in the centrilobular region with portal triaditis.

The Palagarai Parpam 50, 100mg/kg treated group showed minimal inflammation with moderate portal triaditis and their lobular architecture was normal. Silymarin treated group showed normal hepatocytes and their lobular architecture was normal. Since the changes associated with CCl4-induced liver damage are similar to that of acute viral hepatitis. The hepatotoxicity induced by CCl4 is due to its metabolite $CCl_3^{\bullet}$, a free radical that alkylates cellular proteins and other macromolecules with a simultaneous attack on lipids, in the presence of oxygen, to produce lipid peroxides, leading to liver damage.

Amino transferases are present in high concentration in liver, an important class of enzymes linking carbohydrate and amino acid metabolism. Alanine amino transferase and aspartate amino transferase are well known diagnostic indicators of liver disease. In cases of liver damage with hepatocellular lesions and parenchymal cell necrosis, these marker enzymes are released from the damaged tissues into the blood stream. Alkaline phosphatase is a membrane bound enzyme and its elevations in plasma indicate membrane disruption in the organ. Alkaline phosphatases, although not a liver specific enzymes, the liver is the major source of this enzyme.

The level of this enzyme increases in cholestasis. Hepatotoxicity is characterized by cirrhotic liver condition which in turn increased the bilirubin release. Thus, In conclusion, the present study, the activities of these enzymes, total bilirubin were found to increase in the hepatotoxic animals, and were significantly reduced in groups of Palagarai Parpam administered rats as compared to that of toxicant rats. The effect was more pronounced with these dose levels of Palagarai Parpam confirms the dose dependent hepatoprotective action in rats.

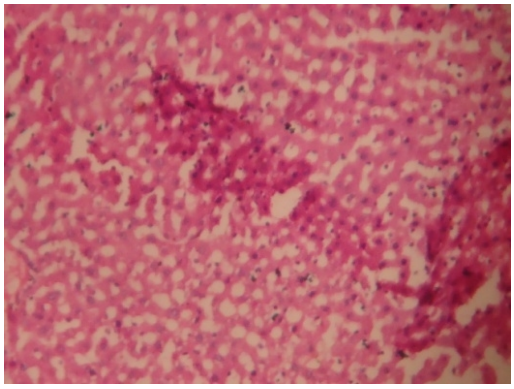
Evaluation of antioxidant activity of Palagarai Parpam by beta-carotene bleaching method

The rapid evaluation of antioxidant activity of Palagarai Parpam was determined according to the beta-carotene bleaching method. In this procedure the Palagarai Parpam, Vit.E were applied on TLC plates and after developing with a suitable solvent system, plates were sprayed with a betacarotene solution and exposed to daylight until discoloring of the background (6h.) The active compounds were seen as orange color on the plate. Vit.E was used as positive control. Palagarai Parpam was showed strong antioxidant activity. The same experiment was repeated twice for confirmation. The developed TLC plate after spraying with the reagent of beta-carotene showed no discoloration of the background after 6 hours.

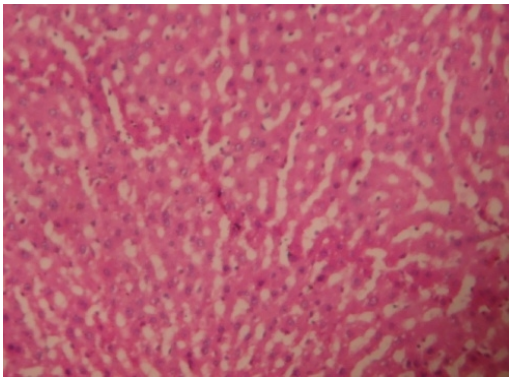
PHARMACOLOGY

HISTOPATHOLOGY

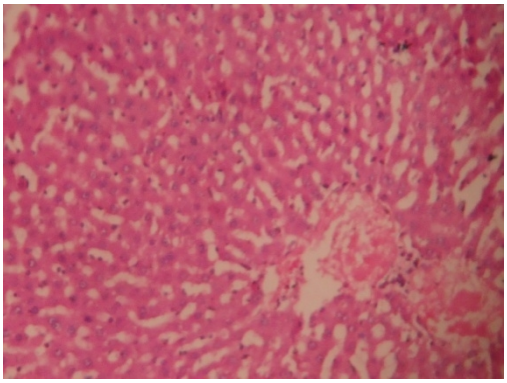
Control



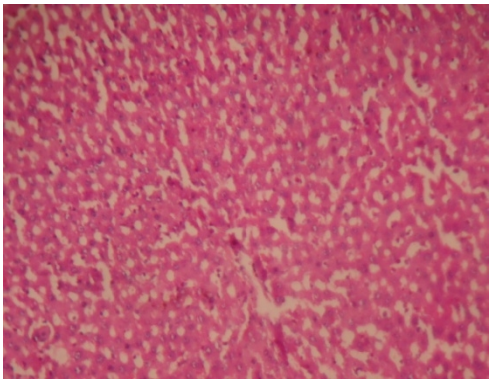
Normal



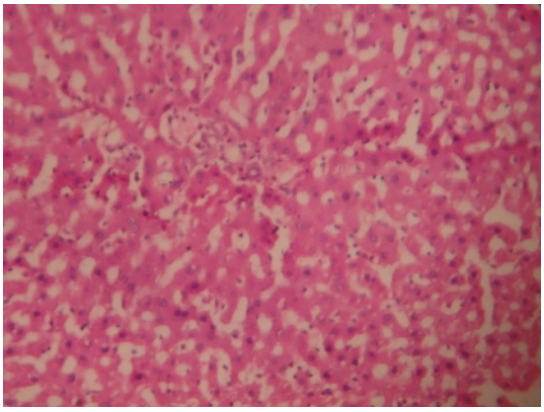
50mg



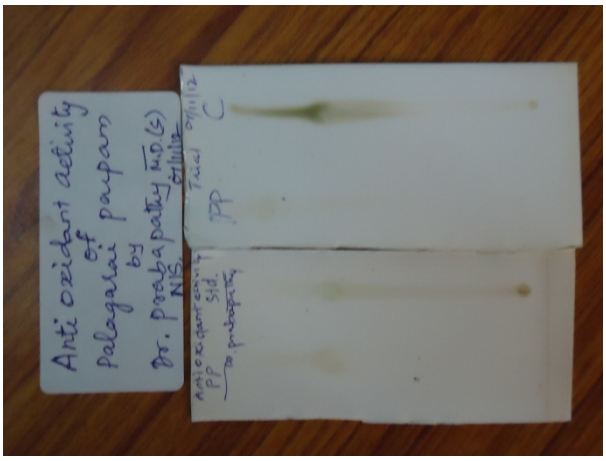
100mg

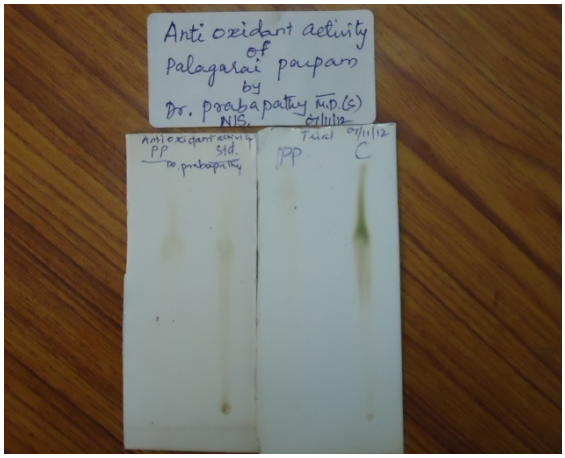


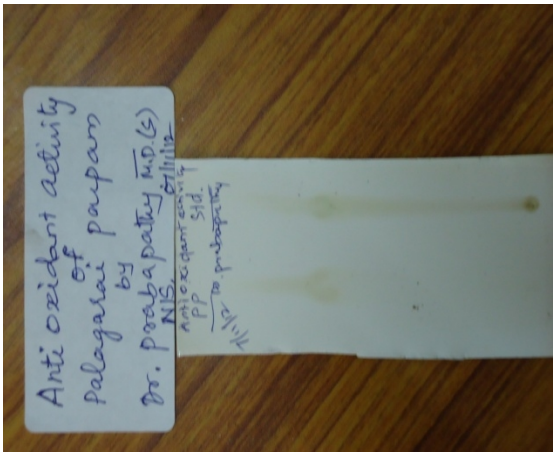
Standard

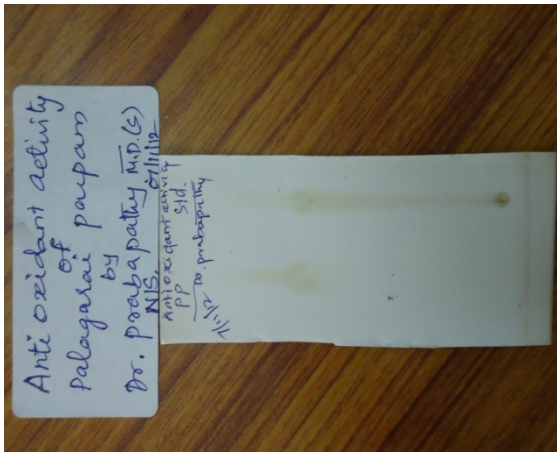


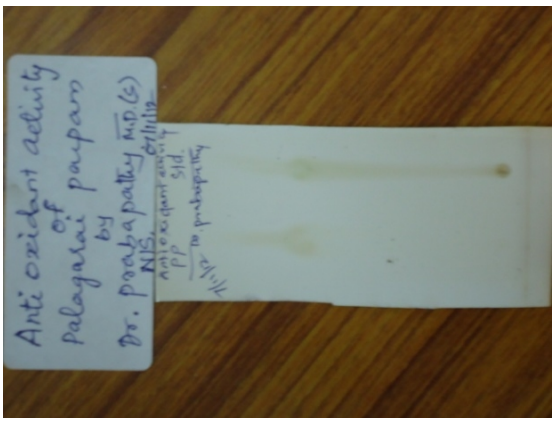
ANTIOXIDANT ACTIVITY

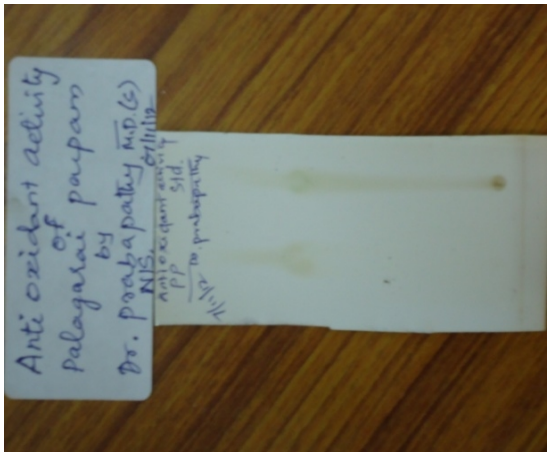












DISEASE ASPECT

SIDDHA ASPECT

கல்லீரல் நோய்கள்[5]

வேறுபெயர்கள் :

வலப்பாட்டீரல் நோய், மாந்தக் கட்டி, கல்மாந்தம், யக்குதம் எனப் பெயர்களுண்டு.

இயல்பு :

இயற்கையாக மார்பின் கீழ் வலப்பக்கம் உள்ள வலப்பாட்டீரல், அப்பக்கமுள்ள கடைவிலா எலும்பு வரையிலும் நிற்காமல் தன்னளவில் மேலுங்கீழுமாகப் பெருத்துக் கொண்டே வருவதும், தன் இயற்கைத் தொழிலை இழப்பதும், அல்லது அளவில் மிகச் சிறுத்துக் கொண்டே வந்து பல நோய்களையும் துணையாக்குவதுமான இயல்பைப் பெறும் நோயாம்.

நோய் வரும் வழி :

1. மிகுந்த அளவில் உணவையுண்ணல், உடற்கொவ்வாப் பொருள்களைக் கொள்ளல்,
2. கள், சாராயம், முதலிய மயக்கத்தைத் தரும் குடி வகைகளை அளவு கடந்து குடித்தல்,
3. பெண்களின் கூட்டால் வரும் நோய் ஆகிவற்றாலும், சுரம் முதலிய நோய்கட்குத் துணையாகவும் வருவதாகும்.

முற்குறி :

வாய் கைத்தல், சுவையின்மை, வாய்நீருறல், பசியின்மை, உண்ட உணவு செரியாமை, காலையில் பித்தமாக வாந்தியாதல், முகம் சுருங்கி முக எலும்பு எடுத்துக்காட்டல், கை கால் சும்பி வயிறு நாட்குநாள் பெருத்துக் கொண்டே வருதல், அடிக்கடி சுரங்க காய்தல் என்னும் குறிகளைக் காட்டி வலப் பாட்டீரல் பெருத்துக் கொண்டே வரும்.

நோய் எண் :

குற்ற அளவாக மூன்று வகைப்படும். அவை வளிக் கல்லீரல் நோய், அழல் கல்லீரல் நோய், ஐயக் கல்லீரல் நோயாம்.

வளிக் கல்லீரல் நோய் :

1. உடலின் கண் வளிக் குற்றம் பெருகித் தனக்குத் துணையாக அழல் குற்றத்தையுங் கூட்டி, சுரத்தையுண்டாக்கி, உடலை நாளுக்கு நாள், இளைக்கச் செய்யும்
2. வயிறு பெருத்துக் கொண்டே வருதல், இரச நாளங்களின் முடிச்சுகள் கனத்துத் தொடை இடுக்குகளிலும், அக்குள், வயிறு, கழுத்து, மார்பு இவற்றில் தோன்றும்.
3. நாட் செல்லச் செல்லப் படுக்கையிலிருத்தித் துன்புறுத்தும்.
4. நோய் முதிருங்காலத்தில் உடலின் குருதி குறைந்து உடல் வெளுத்துக் கால், கை, வயிறு ஆகியவைகளும் வீங்கிக் காணும்.

அழல் கல்லீரல் நோய் :

1. கல்லீரலின் இயற்கைச் செயலுங்குன்றிப் பித்துநீரை உடல் முழுமையும் வீசி உடலை மஞ்சள் நிறமாக்கும்.
2. வாய் கைத்தல், பித்து வாந்தியாதல், முகஞ் சுருங்கல், கை கால் வீங்கல், குருதியின் வன்மையின்மையால் உடல் வெளுத்துக்கனால் என்னுங் குறிகளைக் காட்டும்.
3. பெரு வயிறு நோயையும் பின் தொடரச் செய்யும்.

ஐயக்கல்லீரல் நோய் :

1. கல்லீரல் மிகப் பெருத்து, தொடுகைக்குக் கட்டி முட்டியாகக் காணப்படும்.
2. சிறுநீர் சிவந்து, அளவில் குறைந்து இழியும்
3. மஞ்சள் (காமாலை) நோய், உடல் வீங்குதல், உடல் வெளுத்தல்

தீரும் தீராதவை :

வளிக்குற்றத்தால் வரும் நோயும் ஐயக்குற்றத்தால் வருநோயும் தீராதாம்.

பொதுக்குணங்கள் :

1. உணவில் வெறுப்பு, நாச்சுவையறியாமை, உண்ணினும் வாய் குமட்டி குமட்டி வாந்தியாதல், காலையில் எழுந்த உடனே வாய் குமட்டிப் பித்துநீராக வாந்தியெடுத்தல், உண்ட உணவும் சரிவர செரியாதிருத்தல்.
2. செரிப்பினும் உடற்கு ஊட்டமளிக்காமல் உடலை இளைக்கச் செய்தல், முகஞ்சுருங்குதல், தாடை எலும்பு வெளித் தோன்றல்
3. கை, கால் சும்பி, உடல் வெளுத்து வயிறு பெருக்கத் தொடங்கும். கல்லீரலும் நாளுக்குநாள் பெருத்துக் கொண்டே வரும்.

குற்ற வேறுபாடு :

அழல் (பித்த) குற்றத்தாலெழுந்து தனக்குத் துணையாக மற்ற இரு குற்றங்களைக் கூட்டிக்கொண்டு பரவுகாலின் செயலைக் கெடுத்து வருநோயாம்.

உணவு :

பசித்தீ மிகவும் கெடுமாதலால் கடின உணவுகள் எளிதில் செரியாதாகையின் இருமுறை வடித்த சோறு அல்லது கஞ்சி வகைகளில் ஏதேனும் ஒன்றைக் கொடுக்கலாம். கத்தரிப் பிஞ்ச, பப்பாளிக்காய், முருங்கைப் பிஞ்சு, அவரைப் பிஞ்சு போன்றவையும் ஆட்டுக்கறி, காடை, கௌதாரி, உடும்பு, நத்தை நண்டு இவற்றை வழங்கலாம். கீரை வகைகளுள் முள்ளங்கி சிறு கீரை, முள்ளிக்கீரை, பசலைக்கீரை ஆகிய இவற்றைத் துவட்டியுண்ணலாம்.

கஞ்சி வகைகளையும் வெள்ளாட்டின் கறி, காடை கௌதாரி இவற்றின் குடிநீர் (சூப்பு), சிறுகீரை, பொன்னாங்காணி, கரிசாலை, கத்திரிப்பிஞ்சு, முருங்கைப் பிஞ்சு, பீர்க்கு, புடலை, வெள்ளைப்பூண்டு, வெங்காயம், பப்பாளிப் பிஞ்ச இவற்றை உண்ணலாம்.

MODERN ASPECT

Liver disease[1]

Liver disease is invariably applied to many disease and disorder, which cause the liver to function improperly and may lead even to stop functioning . Abnormal pain, yellowing of skin or eye (jaundice) or abnormal result of liver function test are the suggestion features of liver disease[1].

Usually liver disease classified as hepatocellular diseases, cholestastics (obstructive) or mixed.[21]

Hepatocellular diseases are viral hepatitis, alcoholic liver disease. In hepatocellular diseases the features of liver injury, inflammation, necrosis are predominant.

Cholestastic diseases are gallstone, malignant obstruction, primary biliary cirrhosis, some drug -induced liver diseases. In this disease features of inhibition of bile flow is predominate.

Mixed pattern liver diseases are cholestastic forms of viral hepatitis, many drug induced liver diseases. In mixed pattern both hepatocellular and cholestastic injury present. Liver diseases are classified as acute and chronic liver diseases.

Acute liver diseases:[22]

Symptoms :

This may asymptomatic and anicteric, symptomatic diseases, which is often viral, produces symptoms of malaise, anorexia, and fever. Jaundice may appear as illness progresses.

Signs:

- Jaundice
- Enlarged liver
- Pale stools (cholestastic)
- Liver palms (severe acute liver diseases)
- Dark urine
- Spider navi

Chronic liver diseases

Symptoms:

Patients may be asymptomatic or complain of non- specific symptoms, particularly fatigue

Specific symptoms:

- Right hypochondric pain
- Abdominal distention
- Ankle swelling
- Pruritus
- Breast swelling
- Loss of libido
- Amenorrhoea
- Confusion and drowsiness

Signs

General sign:

- ±Fever
- ±Jaundice
- Loss of hair

Compensated Sign:

- Xanthelamas
- parotid enlargement
- spider navi
- liver (small or large)
- spleenomegally
- gynaecomastia
- liver palms
- scratch marks
- purpura
- testicular atrophy

De compensated sign

- Neurological i.e. disorientation
- Drowsy
- Hepatic flap
- Coma
- Ascites
- Dilated veins on abdomen
- ± Oedema

CLINICAL STUDY

STUDY DESIGN & CONDUCT OF STUDY.

Study place : NIS (OPD,IPD)

Period : 12 months

Sample size : 20 patients (both sex)

Weight : 40-85 kg

Dose : 130 mg with ghee or butter

Duration : 30 days

SUBJECT SELECTION:

Patients reporting at OPD of Ayothidoss Pandithar hospital with symptoms of inclusion criteria were subjected to screening test & documented using screening proforma.

Inclusion criteria:

1. Age :20-60 years
2. Sex :both male and female
3. Weight :40-85 kg
4. Patient having symptoms of
   - Malaise
   - Anorexia
   - nausea
   - vomiting
   - Jaundice
   - Upper abdominal pain
5. Patient who are willing to provide blood for lab investigation.
6. Patient willing to sign the informed consent stating that he/she will conscientiously stick to the treatment during 30 days but can opt out of the trial of his/her own conscious discretion.

Exclusion criteria:

- Cardiac disease
- Hepatic failure
- Pregnancy and lactation
- Any other serious illness

Withdrawal criteria:

1. Development of any adverse reaction
2. Occurance of any other serious illness

TESTS AND ASSESMENTS

A. Clinical assessment

Siddha assessment

B. Laboratory Investigations

1. Routine investigations

2. Specific investigations

A.CLINICAL ASSESSMENT

- Malaise
- Anorexia
- nausea
- vomiting
- Jaundice
- Upper abdominal pain

5. Patient willing to provide **SIDDHA PARAMETERS**

1. Naa
2. Niram
3. Mozhi
4. Vizhi
5. Sparisam
6. Naadi
7. Malam
8. Moothiram –Neerkuri and Neikuri

ROUTINE INVESTIGATION

BLOOD

- Hb (gm/dl) Total WBC Count(Cells/cumm) ,
- DC - (Polymorphs (%), Lymphocytes (%) Eosinophils(%) , Monocytes(%), Basophils (%),
- Total RBC count (Million cells / cu mm),
- ESR (mm/hr)
- Blood glucose (mg/dl) (Fasting, Post Prandial or Random)

LIPID PROFILE

- Serum cholesterol (mg/dl), HDL cholesterol (mg/dl), LDL cholesterol (mg/dl)- VLDL cholesterol (mg/dl), Serum triglycerides (mg/dl).

KIDNEY FUNCTION TESTS

- Blood urea(mg/dl), Serum Creatinine (mg/dl)

LIVER FUNCTION TESTS

- Serum total bilirubin (mg/dl) , Serum Direct bilirubin (mg/dl) , Serum Indirect bilirubin (mg/dl), Serum Alkaline phosphate (u/l) , SGOT (u/l), SGPT (u/l), Serum Total Protein (g/dl) , Serum Albumin(g/dl), Serum Globulin(g/dl), Serum Calcium (mg/dl), Serum Phosphorous (mg/dl), Serum Uric Acid (mg/dl).

URINE

Urine sugar (F) & (PP) or (R), Albumin, Deposits Bile salts,Bile pigments,Urobilinogen.

MOTION

- Ova, Cyst, Occult blood.

OTHER INVESTIGATION

- USG Abdome

Specific investigations:

Liver Function Test

Urine:

Bile salts

Bile pigments

STUDY ENROLLMENT:

- In this clinical study, patients reporting at the OPD with the clinical symptoms of malaise, anorexia ,nausea ,vomiting ,jaundice ,upper abdomen pain etc will be examined clinically for enrolling in the study based on the inclusion and exclusion criteria.
- The patients who are to be enrolled would be informed (Form IV C) about the study, trial drug, possible outcomes and the objectives of the study in the language and terms understandable to them.
- After ascertaining the patient's willingness, informed consent would be obtained in writing from them in the consent form (Form IV-A).
- All these patients will be given unique registration card in which patients' Registration number of the study, Address, Phone number and Doctors phone number etc. will be given, so as to report easily should any complications arise.

- Complete clinical history, complaints and duration, examination findings-- all would be recorded symptoms in the prescribed Proforma in the history and clinical assessment forms separately. Screening Form- I will be filled up; Form I-A, Form –II and Form –III will be used for recording the patients' history, clinical examination of symptoms and signs and laboratory investigations respectively.
- Patients would be advised to take the trial drug and appropriate dietary advice (Form IV-D) would be given according to the patients' perfect understanding.

CONDUCT OF THE STUDY:

- Liver disease patient who satisfying the inclusion criteria will be admitted to the trial.
- Patient informed consent will be obtained
- For OP patients ,they should visit the hospital once in 7 days. At each clinical visit clinical assessment is done and prognosis is noted.
- For IP patients clinical assessment is daily and prognosis is noted.
- Laboratory investigations are done before the trial started and at end of the trial for both

OP & IP patients

CLINICAL OBSERVATION:

From the clinical study 75% of patient relieved from upper abdomen , relieved from vomiting,71% of patients relieved from nausea,69 % of patients relieved from anorexia,

63 % of patients relieved from malaise,65% of patients relieved from jaundice and no adverse effects were observed during trial period**.**

DISCUSSION

The principle aim of this study was to assess the pre-clinical safety and efficacy and to evaluate the therapeutic efficacy of the drug Palagarai Parpam in the management of Kalleral Noi.

As per Siddha text, in Kalleral noi, Pitham humours was deranged and then Vatham, Kabham Thathu were deranged. Pitham is responsible for the functioning of digestion , maintenance of the blood environment (Udal Thathu - Senneer), appetite. Hence Pitha thathu when deranged produces symptoms like nausea, vomiting, malaise, and abdomen pain.[23]

The trial drug Palagarai Parpam possess Kaippu suvai and Kaarppu veeryam, hence it balances the deranged Pitha Kutram. In addition to this it also have sedative activity which exert the soothing effect.[10] As per Siddha text Kaippu Suvai detoxified the toxins,[24] nowadays toxins are major cause of Kalleral noi (liver disease). Hence administration of the trial drug Palagarai Parpam was effective in the management of Kalleral noi.

Biochemical analysis:

Biochemical analysis of the drug Palagarai Parpam reveals the presence of Sodium, Calcium, Phosphate , Iron, and Alkaloids and Chloride.

Calcium:

Calcium malabsorption has been demonstrated in primary biliary cirrhosis (PBC) and in hepato-cellular disease; this has been attributed to decreased intestinal absorption of vitamin D.

Sodium:[26]

Important for acid-base balance.

Required for normal muscle irritability and cell permeability.

Sodium deficiency causes muscle cramps.

Sodium reduces prostaglandin synthesis.

Role of **Alkaloid** in treating Kalleral noi (liver disease):

It possesses anti-oxidant property and causes induction of anti-oxidant enzymes like Superoxide dismutase and reduces glutathione & catalase. Also stimulates heme oxygenase – 1 activity.

The preliminary phytoconstituents screening of the trial drug Palagarai Parpam was done by using AAS The result shows the presence of Zinc, Calcium, Magnesium.

Magnesium:[27]

It has antioxidant properties and is need to activate as number of enzymes, helps the body to absorb vitamin B, and vitamin E. Magnesium inhibit lipid peroxidation.

Calcium:

Importance of **calcium** supplement in liver disease

Calcium malabsorption has been demonstrated in primary biliary cirrhosis (PBC) and in hepato-cellular disease; this has been attributed to decreased intestinal absorption of vitamin D

Zinc:

Zinc supplement Prevent Liver disease.[28] Zinc plays an important role in the protection of cell membrane integrity and may be protective against free radical injury. Zinc is needed for the functions of over 100 enzymes. It is essential for DNA, RNA and protein synthesis and, as such, is important for cell division. Recent reports indicate that in human subjects thymopoietin may be zinc dependent and in animal studies somatomedin may be affected adversely due to dietary zinc restriction.

The clinical features of cirrhosis of the liver, poor appetite, susceptibility to infections and enhanced sensitivity to drugs, may be related to conditioned deficiency of zinc.*

AAS analysis of the drug Palagarai Parpam reveals the presence of Calcium, Zinc.

Toxicologicological studies

The results indicated the toxic effect upto 500mg/kg of Palagarai Parpam *treated* via oral route over a period of 28 days. So, it can be concluded that the Palagarai Parpam can be prescribed for long term therapeutic use in human with the necessary dosage reduction and can be used up to 250mg/kg. body weight p.o.

***(The role of zinc in gastrointestinal and liver disease.)**Prasad AS)

Pharmacological Studies

In conclusion, the present study, the activities of these enzymes, total bilirubin were found to increase in the hepatotoxic animals, and were significantly reduced in groups of Palagarai Parpam administered rats as compared to that of toxicant rats. The effect was more pronounced with these dose levels of Palagarai Parpam confirms the dose dependent hepatoprotective action in rats.

Clinical observation:

From the clinical study 75% of patient relieved from upper abdomen , relieved from vomiting,71% of patients relieved from nausea,69 % of patients relieved from anorexia,

63 % of patients relieved from malaise,65% of patients relieved from jaundice and no adverse effects were observed during trial period**.**

Total bilirubin level significantly reduced for 72% of patients.

SGOT level reduced significantly for 83% of patients.

SGPT level reduced significantly 83% of patients.

Bio-statistics:

Statistically, the paired 't' test shows statistical significance for the symptoms before and after the treatment.($p<0.0001$).

SUMMARY

The literary evidence strongly supports the hepatoprotective activity of Palagarai Parpam. The literary evidence from Kannusamy parambarai Vaithiyam strongly support the hepatoprotective activity of the drug.

The qualitative and quantitative analyses were done at Biochemistry lab, NIS and SRU, Chennai respectively. The biochemical analysis of the drug reveals the presence of zinc, magnesium, calcium, alkaloid, amino acids. The results ensure the Hepatoprotective and Antioxidant activity of the Palagarai Parpam was due to the presence of active phytoconstituents of the drug.

The pre-clinical evaluation (acute & repeated oral toxicity study) of the drug was carried out as per OECD guideline in Vels college of pharmacy, Chennai. The result shows safety of the drug for human administration.

The Preclinical Pharmacological study was carried out in animal model in Vels college of pharmacy, Chennai. The result shows that the drug has significant hepatoprotective activity.

As per the Siddha literature and modern science reviews and research articles, the trial drug has potent Hepatoprotective effect.20 Patients were recruited for clinical trial. The trial drug hepatoprotective activity at the dose of 130 mg, bid was given to the patient for 7 days and patients were asked to visit op once in 7 days for 30 days. Clinical assessment and prognosis was noted at each visit.

From the clinical study 75% of patient relieved from upper abdomen , relieved from vomiting,71% of patients relieved from nausea,69 % of patients relieved from anorexia,63 % of patients relieved from malaise,65% of patients relieved from jaundice and no adverse effects were observed during trial period.

Total bilirubin level significantly reduced for 72% of patients.

SGOT level reduced significantly for 83% of patients.

SGPT level reduced significantly for 83% of patients

The drug Palagarai Parpam. has

- Hepatoprotective activity.and Antioxidant Activity.
- No side effects
- No undoing effects
- Encouraging clinical resultsFrom the clinical and the statistical analysis, it is proved that the drug Palagarai Parpam is statistically significant on Hepatoprotective activity on Kalleral noi.

CONCLUSION

- The literature and research journal review of the herbo mineral drug shows that it has hepato-protective activity.

- The safety studies (acute toxicity and sub-acute l toxicity) studies conducted revealed that the trial drug palagarai parpam is safe. There were no abnormalities found in blood investigation and histo-pathological examination. Hence it can be reasonably assumed that the drug is safe for human use.

- The pharmacological study conducted in animal model shows significant hepato-protective activity.

- Clinical study revealed the therapeutic efficacy of the trial drug by showing, reduction in serum enzyme markers SGOT, SGPT level ,Total bilirubin level significantly. There was improvement in other clinical symptoms before and after treatment.

- There were no adverse reactions complained during the clinical trial.

Hence, the drug Palagarai Parpam can be used in the management of Kalleral noi

INTRODUCTION

Soolai (Osteoarthritis-OA) is characterized by focal loss of articular cartilage and proliferation and remodelling of bones around the joint to form osteopyte. Inflammation can be a feature of Osteoarthriti.[29]

Osteoarthritis (OA) represents an imbalance in destruction and synthetic process of the cartilage lead to the erosion, decreased concentration and viscosity of the synovial fluid, decreased lubricating and cushioning properties. There is also an underlying inflammation of the synovium as well as damage in the subchondral bone.

Several factors predispose to the disease and accelerate its progression. These includes pre-existing joint diseases, obesity, hyper mobility, orthopedic deformity, endocrine disorder like diabetes mellitus, hyperparathyroidism. Overuse of joints and adoption of unusual posture for long periods predispose to the condition.

Among the elderly, Knee Osteoarthritis is the leading cause of disability in developed country.[21]

Osteoarthritis (OA) affects 10-15% of world population.[30] About 5.7% of Population of India has Osteoarthritis (OA).[31] In general prevalence of osteoarthritis increases with age. 80% of people affected by 40 years. More than 50% have bilateral Osteoarthritis.[32] Women have great tendency than men.[32] Genetic tendency in knee Osteoarthritis is twice as that of osteoarthritis hip.

Chithiramoola Rasayanam is mentioned in Siddha literature, Pulipanivaithiyam 500 for Soolai. Soolai generally means deep seated pain.[33] Such a pain is experienced in arthritis. As per Siddha text Soolai means Soolam ennum karuviyal kuthungal undakum valiyaiyottha noi.[6]

Rasayana means a medicine, which prolongs life and neutralise deranged Vatham, Pitham, Kapham. Soolai is caused by deranged Vatham[33]. The sage Therayar has quoted that "Nedu vatha sarvathumandri Soolai varathu".[6]

Hence the author selected Chithiramoola Rasayanam for Anti-inflammatory activity in the management of Soolai.

AIM:

To evaluate the safety and efficacy of Chithiramoola Rasayanam for anti-inflammatory activity in the management of Soolai (Osteoarthritis).[34]

OBJECTIVE:

Primary objective:

To evaluate the Anti-inflammatory activity of "Chithiramoola Rasayanam" for Soolai (Osteoarthritis) in preclinical studies.

Secondary objectives:

Biochemical analysis

To evaluate the efficacy of Chithiramoola Rasayanam in the management of Soolai (Osteoarthritis)

High performance thin layer chromotography

MATERIALS AND METHODS

STANDARD OPERATIVE PROCEDURE:

COLLECTION AND AUTHENTICATION OF RAW DRUGS:

The raw drug was procured from Ramasamy chetty shop Paris Chennai and authenticated by competent authority of department of Gunapadam, National institute of Siddha.

INGREDIENTS:

Purified Chithiramoolam (Plambago indica)- 30 palam(1050 gm)

Purified Nilappanai (Curculigo orchiodes)- 30 palam (1050gm)

Purified Thannirvittan (Asparagus racemosos)- 30 palam(1050 gm)

Purified Parangichakkai (Smilax china)_30 palam (1050 gm)

Purified Cherangkottai (Semecarpus anacardium)-40 palam(1400 gm)

Honey- 80 palam (2800 gm)

Ghee -80 palam (2800 gm)

Vellai Sarkarai (Saccharum officinarum)-160 palam (5600 gm)

Purification process:

Purification of Chithiramoolam[35]:

The outer covering was powdered and then baked in milk steam for 3 hours and then powdered .

Purification of Nilappanai[36]:

The drug was powdered and baked in milk steam for 3 hours, then dried in sunlight and powdered .

Purification of Thannirvittan[36]:

The drug was powdered and baked in milk steam for 3 hours, then dried in sunlight and powdered .

Purification of Parangichakkai[36]:

The drug was powdered and baked in milk steam for 3 hours, then dried in sunlight and powdered.

Purification of Cherangkottai[37]:

Cut the nose like projection of seed and boiled with cow dung's solution. Repeat the procedure for 7 times.

Method of medicine preparation[35]:

Purified Chithirmoolam, Purified Tannirvittan, Purified Nilappanai, Purified Parangichakkai, Purified Cherangkottai are dried well and pulverized by an electric grinder into fine powder and then was sieved by using a fine silk cloth (vasthra kaayam) and then mixed with sugar, honey and ghee. The outcome of Rasayanam stored in a clean dry airtight glass bottle.

LABELLING:

Name of the preparation	: Chithiramoola Rasayanam
Quantity of the drug	: 70gm
Dose	:5 g bid
Adjuvant or Vehicle	: -
Indication	: Soolai (osteoarthritis)
Date of manufacturing	: The drug was prepared in two batches 5/4/12, 5/8/12
Expiry	:6 months from the date of manufacture.

கொடிவேலி

Plumbago zeylanica

பறங்கிப்பட்டை

Smilax China.Linn

தண்ணீர் விட்டான்

Asparagus racemosus

நிலப்பனை

Curculigo orchioides.Gaertn

சேராங்கொட்டை

Semecarpus anacardium.Linn.

CHITHIRAMOOLA RASAYANAM

REVIEW OF THE LITERATURE

GUNAPADAM ASPECT[6]

கொடிவேலி (Plumbago zeylanica)

வேறுபெயர் : கொடுவேலி, சித்திரமூலம், சித்திரம், வன்னி

பயன்படும் உறுப்பு : வேர், பட்டை

சுவை : கார்ப்பு, விறுவிறுப்பு , **தன்மை:** வெப்பம் ,**பிரிவு** : கார்ப்பு.

செய்கை : முறைவெப்பகற்றி, வியர்வையுண்டாக்கி

பொதுகுணம் :

இதனால், கட்டி, புண், கழலை, வளிநோய், அரையாப்புக்கட்டி, குத்தல், சோபை, மூலரோகம், உதிரக்கட்டு, நீரேற்றம், பெருவயிறு இவைபோம்.

"கட்டிவிர ணங்கிரந்தி கால்கள் அரையாப்புக்
கட்டிக்சூ லைவீக்கங் காழ்மூலம் - முட்டிரத்தக்
கட்டுநீ ரேற்றங் கனத்த பெருவயிறும்
அட்டுங் கொடிவேலி யாம்."

நிலப்பனை (Curculigo orchioides.Gaertn)

வேறுபெயர்: வாராகி, முசலி, தலைத்தாது, நேயம், சித்தி

பயன்படும் உறுப்பு : கிழங்கு, வேர்

சுவை : இனிப்பு ,**தன்மை :** தட்பம் ,**பிரிவு :** இனிப்பு.

செய்கை : உரமாக்கி, அகட்டுவாயகற்றி,

குணம் :

இதனால், நீரிழிவு, வெப்பம், வெண்புள்ளி, கரும்புள்ளி, விலாக்குத்தல், ஒழுக்கு வெள்ளை, கண்நோய்கள் இவை போம். ஆண்மையுண்டாகும்.

"மேக வளனல்தணியும் வெண்குட்டந் தான்விலகும்
போக மிகவுமுறும் பொற்கொடியே! - போகாத
சூலைமே கங்களோடு துன்னுகரும் புள்ளியும் போஞ்
சால நிலப்பனைக்குத் தான்"

தண்ணீர்விட்டான் (Asparagus racemosus Willd)

வேறுபெயர் : தண்ணீர்விட்டான், சதாவேலி, சதாவேரி, நீர்விட்டான்

பயன்படும் உறுப்பு : இலை, கிழங்கு

சுவை : இனிப்பு **தன்மை :** வெப்பம் **பிரிவு :** இனிப்பு

செய்கை : உடலுரமாக்கி, உள்ளழலாற்றி, காமம்பெருக்கி

குணம் :

இதன் கிழங்கு நீரிழிவு, நாட்பட்டசுரம், எலும்புருக்கி நோய், வெந்நீரை அழிக்கும் நோய், வெட்டை, உட்சூடு முதலியவற்றை நீக்கும்.

பறங்கிப்பட்டை (Smilax China.Linn)

வேறுபெயர்: மதுஸ்மிகம், மதுஸ்மீகி, சீனப்பட்டை, பறங்கிசக்கை

பயன்படும் உறுப்பு : கிழங்கு

சுவை : இனிப்பு, **தன்மை :** வெப்பம், **பிரிவு :** இனிப்பு,

செய்கை : உடற்றேற்றி, மேகப்பிணிவிலக்கி, காமம்பெருக்கி, தூய்மையாக்கி

குணம் :

இதனால் நீர்வேட்கை, பற்பல வளிநோய், புண், பிளவை, நீரிழிவு, கடிவிடம், சிரங்கு, மூலமுளை, முடவாதம், குறைநோய், ஐயம், மகரந்தப்புண், வாந்தி இவை நீங்கும். ஆண்மை உண்டாம்.

“தாகம் பலவாதந் தாதுநட்டம் புண்பிளவை
மேகங் கடிகிரந்தி வீழ்மூலந் - தேகமுடன்
குட்டை பகந்தமேற் கொள்வமனம் போம்பறங்கிப்
பட்டையினை யுச்சரித்துப் பார்.”

சேராங்கொட்டை (Semecarpus anacardium.Linn)

வேறுபெயர் : சேங்கொட்டை, வல்லாதி, வல்லாதகி, எரிமுகி, பல்லாதகி, கிட்டாக்கனிக் கொட்டை, நந்திவித்து

பயன்படும் உறுப்புகள் : கொட்டை, பருப்பு

சுவை : கைப்பு, விறுவிறுப்பு, **தன்மை :** வெப்பம், **பிரிவு :** கார்ப்பு.

செய்கை : உடற்றேற்றி, புண்ணாக்கி

குணம் :

"குட்டங் கயரோகங் கொல்லும் விடபாகந்

துட்டந் தருகிருமி சூலையும் போம் - மட்டலருங்

கூந்தன்மயி லேகிரந்திக் கூட்டம் போஞ் செங்கையில்

ஏந்துசேங் கொட்டைதனை யே."

இது பெருநோய், இளைப்பு நோய், நஞ்சுகள், சூலை இவைகளைப் போக்கடிக்கும். மேலும் திமிர்ப்பட்டை, கருப்புப் படை, வெண்படை (வெண் குட்டம்), தீராக்கடி, மூலம், வளி நோய்கள், குன்மம் இவைகளையும் விலக்கும்.

சேரும் மருந்துகள்

சித்திர மூலம் சேரும் மருந்துகள் :

சித்திர மூல நெய் :

அளவு ; கரண்டியளவு

தீரும் நோய்கள் : சூலை, குன்மம்

ஆதாரம் : அகத்திய முனிவர் அருளிச்செய்த வைத்திய ரத்தின சுருக்கம் 360

மதுஸ்மிகி ரசாயணம் :

அளவு : பாக்களவு, இருவேளை

தீரும்நோய்கள் ; சூலை, வாதநோய்கள்

ஆதாரம் : அகத்திய முனிவர் அருளிச்செய்த வைத்திய ரத்தின சுருக்கம் 360, ப. 40

சூலைக் கியாழம் :

தீரும்நோய்கள் : சூலை

ஆதாரம் : கண்ணுசாமி பரம்பரை வைத்தியம் ப. 71

வாத சுர கியாழம் :

தீரும்நோய்கள் : கை கால் குடைச்சல், சூலை

ஆதாரம் ; அகத்தியர் முனிவர் அருளிச் செய்த வைத்திய ரத்தின சுருக்கம் 360, ப. 111

நிலப்பனை கிழங்கு சேரும் மருந்துகள் :

பறங்கி ரசாயனம் :

அளவு : கழற்சிகொட்டையளவு

தீரும் நோய்கள் : சூலை

ஆதாரம் : கண்ணுசாமி பரம்பரை வைத்தியம் ப. 461

கந்தக ரசாயனம் :

அளவு : கழற்சிகொட்டை அந்தி, சந்தி

தீரும் நோய் : சூலை

ஆதாரம் : கண்ணுசாமி பரம்பரை வைத்தியம்

தண்ணீர்விட்டான் கிழங்கு சேரும் மருந்து :

நரசிம்மலேகியம்

அளவு : 5-10 கிராம்

தீரும்நோய்கள்: வாதம்

வழக்கு: தண்ணீர் விட்டான் கிழங்கின் இரசம், தேன் சேர்த்து குடித்து வர சூலை நீங்கும்

ஆதாரம் : குணபாடம் -முதல் பாகம்.

பறங்கிப்பட்டை சேரும் மருந்துகள் :

மேகவாயுவிற்கு கியாழம்

தீரும்நோய்கள் : குடைச்சல்

ஆதாரம் : கண்ணுசாமி பரம்பரை வைத்தியம் ப. 460

பறங்கி ரசாயனம் :

அளவு : கழற்சி கொட்டையளவு, அந்தி, சந்தி

தீரும் நோய்கள் : சூலை, மேகவாயு

ஆதாரம் : கண்ணுசாமி பரம்பரை வைத்தியம் ப. 461

பறங்கி ரசாயனம் ;

அளவு : வெருகடியளவு

தீரும்நோய்கள் : பதினென் சூலை

ஆதாரம் : போகர் வைத்தியம் 700

சேராங்கொட்டை சேரும் மருந்து

இரசகந்தி மெழுகு :

அளவு : சுண்டைக்காய் அளவு

தீரும் நோய்கள் : வாத சூலை, கால் குடைச்சல்

ஆதாரம் : சித்த வைத்திய திரட்டு

இடிவல்லாதி :

அளவு : சுண்டையளவு

தீரும் நோய்கள் : சூலை

ஆதாரம் : சித்த வைத்திய திரட்டு

BOTANICAL ASPECT[17]

கொடிவேலி

BOTANICAL NAME : Plumbago zeylanica Linn
VERNACULAR NAME:

ENGLISH : Ceylon leadwort

SANSKRIT : Chitraka

HINDI :Chita

TELUNGU :Aagnimatu

MALAYALAM :Thumbu kodiveli

CLASSIFICATION :

Kingdom:Plantae

Order :Caryophyllales

Family :Plumbaginaceae

Genus :Plumbago

Species: *P. zeylanica*

Binomial name: Plumbago zeylanica Linn

BOTANICAL DESCRIPTION:

Perennial, sub-scandent shrub ,160-120cm high. Leaves alternate, ovate, acute, glabraous, entire, stalk short, flower white, in bracteate, often branched, glandular and elongated spikes, 10-30cm long. Capsule oblong, pointed, contained in viscid, glandular, persistent calyx. seeds oblong.

PARTS USED:

Roots, root bark.

PHYSICAL CONSTANTS:

Foreign matter - not more than 30%.

Total ash -not more than 3%

Acid insoluble ash -not more than 1%

Alcohol extractive - not less than 12%

water soluble extractive -not less than 12%

CHEMICAL CONSTITUENTS:

plant contains number of naphthoquinone derivatives viz plumbagin,3-chloroplumbagin , 3,3-plumbagin,plumbaginic acid , zeylinone.

PHARMACOLOGICAL ACTIVITIES:

Antipyretic, Antitumour, uterotonic

நிலப்பனை

BOTANICAL NAME: : Curculigo orchioids

VERNACULAR NAME:

ENGLISH **:**Black musale

SANSKRIT **:**Musale

HINDI : Musli

TELUNGU **:** Nelatatigadda

MALAYALAM **:** Nilappana

CLASSIFICATION

Kingdom:Plantae

Order:Asparagales

Family:Hypoxidaceae

Genus:Curculigo

Species:C. *orchioides*

Binomial name : Curculigo orchioids

BOTANICAL DESCRIPTION:

Perennial,10-35cm high, root stocks stouts, short or elongated, cylindric, fleshy.

PARTS USED: Root stocks

CHEMICAL CONSTITUENTS:

Curculigosaponins, Curculigosides, Palmetic, Oleic, Linolenic, Linoleic, Flavoneglycosides.

PHARMACOLOGICAL ACTIVITIES:

Anti inflammatory, hypo glycaemic, hepatoprotective, flavanone glycoside showed powerful uterine stimulant**.**

தண்ணீர்விட்டான்

BOTANICAL NAME: Asparagus racemosus Willd.

VERNACULAR NAME:

ENGLISH :Wild asparagus

SANSKRIT : Shatavari

HINDI : Satamuli

TELUNGU : Satavari

MALAYALAM : Satavari

CLASSIFICATION

Kingdom : Plantae

clade :Angiosperms

clade : Monocots

Order : Asparagales

Family : Asparagaceae

Subfamily : Asparagoideae

Genus : Asparagus

Species : Asparagus racemosus

Binomial name : Asparagus racemosus

BOTANICAL DESCRIPTION:

Scandant, much branched spinous undershrub with tuberous, ,short root stocks bearing numerous fusiform tuberous root, 30-100cm long and 1-2cm thick.

PARTS USED:

Tuberous root

CHEMICAL CONSTITUENTS:

Sarasapogenin, saponin A4-A7, diosgenin, asparagamine and disaccharide in roots. Isoflavones, racemosol, polysaccharides, mucilage .

PHARMACOLOGICAL ACTIVITIES:

Phagocytic, Anti-dysentry, gastric sedative, diuretic.

பறங்கிப்பட்டை

BOTANICAL NAME: Smilax china Linn

VERNACULAR NAME:

ENGLISH : China root

SANSKRIT : Dwipathru

HINDI :Chobchin

TELUNGU : Galichekkai

MALAYALAM : Chinapaivu

CLASSIFICATION

Kingdom:Plantae

Order:Liliales

Family: Smilacaceae

Genus:Smilax

Species:*S.china*

Binomial name : *Smilax china*

PARTS USED: Root

CHEMICAL CONSTITUENTS:

Fat , sugar, glucoside, saponin, steroidal saponins, phytosterols, triterpenoids (British Herbal Pharmacopoeia, 1983)

Parillin -the antimicrobial and anti-tumoral activities (Bérdy et al., 1982)

Sieboldogenin

PHARMACOLOGICAL ACTIVITIES:

Alterative

Antisyphilitic

Aphrodisiac

Depurative

சேராங்கொட்டை

BOTANICAL NAME: Semicarpus anacardium Linn

VERNACULAR NAME:

ENGLISH :Marking nut

SANSKRIT :Bhallataka-Bijam

HINDI :Bhilawa

TELUNGU :Nallajidi

MALAYALAM : Chera

CLASSIFICATION

Kingdom: Plantae

Order: Asparagales

Family: Asparagaceae

Subfamily Asparagoideae

genus: Asparagus

Species: Asparagus racemosus

Binomial name : Semicarpus anacardium Linn

BOTANICAL DESCRIPTION:

A moderate sized , deciduous tree,12-15m high. leaves simple, 17.5-60cm long and 5-20cm broad, obvate- oblong ,glabrous above , ashy grey or buff and pubescent beneath .Drupes 2-3cm long, obliquely ovoid, smooth, shining, black when ripe, situated on fleshy orange coloured receptacle.

PARTS USED: Fruit

PHYSICAL CONSTANTS:

Total ash- not more than 4%,Acid insoluble ash -not more than0.51%, Alcohol extractive - not less than 11%,water soluble extractive -not less than 5%.

CHEMICAL CONSTITUENTS:

Bhilawanol, anacardic acid, biflavonoids, essential amino acid-arginine, leucine, lysine.

PHARMACOLOGICAL ACTIVITIES:

Anti-inflammatory, Analgesic, Anti-arthritic, Immunomodulatory, Hypocholesterolemic.

SUPPORTIVE JOURNALS:

Plumbago zeylanica:

The pharmacological results of our current studies revealed that Plumbagin elicited significant anti-inflammatory activities in the carrageenan model in both prophylactic and therapeutic schemes. In present study ,the first evidence showing the anti-inflammatory and analgesic effects of **plumbagin** in vivo through **inhibition of NF-κB activation***.

- The pharmacological results of our current studies revealed that Plumbagin elicited significant anti-inflammatory activities in the carrageenan model in both prophylactic and therapeutic schemes.
- keien ko (1931) find plumbagin **stimulate CNS** it in **small dose** but large dose paralysis occurs
- Plumbagin reduce obesity.[14]

Curculigo orchioides

The root tubers of *Curculigo orchioides* are a potent source of antioxidative phenolic compounds that counteract with ROS responsible for delayed wound healing, and speed up wound healing mechanism. The root tubers of *Curculigo orchioides* significantly increased the level of SOD, NO and decreased LPO in granuloma tissue of diabetic Mice.**

Asparagus racemosus:

It have rich potassium that intervenes in the elemination of corporal liquid and in other very interesting process as **bony calcification.*****

The enzyme activity is also restored due to the presence of enriched therapeutic phytoconstituents which improve the indices of oxidative stress related to aging (Velavan and Begum, 2007a).

***Anti-inflammatory and analgesic effect of plumbagin through inhibition of nuclear factor-kappa B activation**
PEI LUO , YUEN FAN WONG , LIN GE, ZHI FENG ZHANG, YUAN LIU, LIANG IU, HUA ZHOU Centre for Cancer and Inflammation Research, School of Chinese Medicine, Hong Kong Baptist University, Kowloon Tong, Hong Kong (P.L., Y.F.W., L.G., L.L., H.Z.) Ethnic Pharmaceutical Institute, Southwest University for Nationalities, Chengdu, Sichuan Province, P.R. China (Z.F.Z., Y.L.)

**Wound healing activity of standardized extract of *Curculigo orchioides* Gaertn in streptozotocin-induced diabetic mice (Pubmed)

*(www.botanical-online.com)

Semecarpus anacardium*

Ramprasathet *et al.* investigated the Anti-inflammatory effects of Semecarpus anacardium nut extract on developing and developed adjuvant arthritis. *Semecarpus anacardium* significantly decreased the carrageenan-induced paw edema and cotton pellet granuloma. These results indicate the potent Anti-inflammatory effect and therapeutic efficacy of *Semecarpus anacardium* Linn. Nut extract against all phases of inflammation is comparable to that of indomethacin.

Salvem *et al.* investigated that ethyl acetate extract of *Semecarpus anacardium* led to the isolation of major active principle, tetrahydroamentoflavone (THA), a biflavonoid. The *in vitro* cyclooxygenase (COX-1)-catalyzed prostaglandin biosynthesis assay of THA gave an IC50 value of 29.5 μM (COX-1) and 40.5% inhibition at 100 g/mL (COX-2). The *in vivo* carrageenan-induced paw edema assay resulted in dose-dependent Anti-inflammatory effect of THA and the activity was comparable to that of ibuprofen.

Bhitre *et al.* prepared the methanolic, ethanolic, chloroform, ethyl acetate and petroleum ether extracts of fruits of Semecarpus anacardium and tested to study the Anti-inflammatory activity using the technique of carrageenan-induced paw edema in albino rats. The extract showed significant Anti-inflammatory activity comparable to the reference standard aspirin.

Smilax china

The aqueous extract of Smilax china is evaluated for the inhibition of prostaglandin production (for COX-2 inhibitions) in lipopolysaccharide (LPS)-induced mouse macrophage cells. The result showed that both COX-2 activity and COX expression were inhibited by the extract***

It also exhibited significant inhibition of carrageenan-induced hind paw oedema at the doses of 10 and 50mg/kg. Computational molecular docking showed its molecular interaction with important amino acid residues in the catalytic site of lipoxygenase, revealing its potential binding mode at molecular level

**** A review of Semecarpus anacardium***

*****Anti-inflammatory activities of Sieboldogenin from Smilax china Linn.: Experimental and computational studies. Inamullah Khan, Muhammad Nisar, Farooq Ebad, Said Nadeem, Muhammad Saeed, Haroon Khan, Samiullah, Fazli Khuda, Nasiara Karim, Zia Ahmad Department of Pharmacy, University of Peshawar, Peshawar 25120, Pakistan.***

******Journal of Ethnopharmacology (impact factor: 3.01). 11/2008; DOI:10.1016/j.jep.2008.10.009***.

PHYSICAL PROPERTIES

Materials and Methods

The Physical properties of Chithiramoola Rasayanam were analysed in the following procedure done in Sri Ramachandra University.

pH at 10% of aqueous solution:

Five grams of Chithiramoola Rasayanam was weighed accurately and placed in clear 100 ml beaker. Then 50 ml of distilled water was added to it and dissolved well. After 30 minutes it was then applied in to pH meter at standard buffer solution of 4.0,7.0,9.2.

Ash Values

The Ash values measure of the inorganic constituents present in the raw drug. A high ash content explains its unsuitable nature to be used as a drug .

Total Ash

A little of extract was taken in a silica crucible previously ignited, cooled and weighed. It was incinerated by gradually increasing the heat not exceeding dull red heat (450°C) until free from carbon, cooled and weighed. The percentage of ash was calculated with reference to air- dried drug. The procedure was repeated to get the constant weight.

Water soluble ash

The total ash was boiled with 25 ml water and filtered through ash less filter paper (Whatmann 4.1). It was followed by washing with hot water .The filter paper was dried and ignited in the silica crucible, cooled and the water insoluble ash was weighed. The water-soluble ash can be calculated by subtracting the water insoluble ash from the total ash.

Acid insoluble ash

The total ash obtained was boiled for 5 minutes with 25 ml of (10% w/v) dilute hydrochloric acid and filtering through ash less filter paper (Whatmann 4.1). The filter paper was ignited in the silica crucible, cooled and insoluble ash was weighed.

HIGH PERFORMANCE THIN LAYER CHROMATOGRAPHY

Significance of HPTLC fingerprinting in Standardisation

Standardisation of traditional medicine has become mandatory in the present national and international scientific scenario, as they have to stand competing with stringent regulatory methods and also clinically. HPTLC is one of the versatile chromatographic methods presently available for the rapid analysis of herbal drugs due to several reasons. Firstly the time required for the demonstration of the most of the characteristic constituents of a drug is very quick and short. Secondly, in addition to qualitative detection, HPTLC also provides semi-quantitative information on the major active constituents of a drug, thus enabling an assessment of drug quality. Thirdly the fingerprint obtained is suitable for monitoring the identity and purity of drugs and for detecting adulteration and substitution. Hence in order to check the identity, purity and standardise the quantity of active principles in the herbal extracts a HPTLC fingerprint used in the formulation has been obtained.

The distribution of phyto-constituents in a plant depends on various factors such as soil, time of collection period of storage, etc. So, it is necessary to standardize the extract being used for pharmacological studies. HPTLC serves as a convenient tool for finding out the distribution pattern of phyto constituents which is unique to each plant. The HPTLC finger-printing profile establishes the identity and purity of the raw drug being used. It helps in the authentification of the plant material.

Chromatographic Conditions

The finger printing has been done using the following chromatographic conditions. Chromatography was performed on a10x10 cm pre activated HPTLC silica gel 60F 254 plate. Samples were applied to the plate as 6mm wide band with an automatic TLC applicator Linomat 5 with N2 flow (CAMAG, Switzerland), 8mm from

the bottom. Densitometric scanning was performed on CAMAG scanner III.The plates were pre-washed by methanol and activated at 60^0 C for 5 minutes prior to chromatography. The slit dimension was kept at 5 minutes x 0.45 minutes and 20 minutes scanning speed was employed. The mobile phase was chosen after running each plant in different mobile phases of varying polarity (Toluene, Toluene: Ethyl acetate and Ethyl acetate: Methanol) and 10 ml of mobile phase was used per chromatography. Linear ascending development was carried out in 20 cm x 10-em twin glass chamber saturated with the mobile phase.

Chromatographic Analysis

The hydro alcoholic extracts of the plants have been prepared at a concentration of 10 mg/10 ml in alcohol and were spotted using CAMAG Linomat 5 applicator. The method was optimized by selecting appropriate mobile phase for respective plant extracts and developed in a twin trough chamber, 20 x 10 cm at 25°C. The plates were dried by hair dryer. The developed plates were scanned at appropriate wavelength using CAMAG TLC scanner 3 and photo-documented using CAMAG REPROSTAR 3.

Inferences

The finger-printing profile establishes the identity and purity of the raw drug being used. It helps in the authentication of the respective plant material. The finger-printing pattern is characteristic of each plant material used for pharmacological studies. The pattern clearly displays the variation from plant to plant.

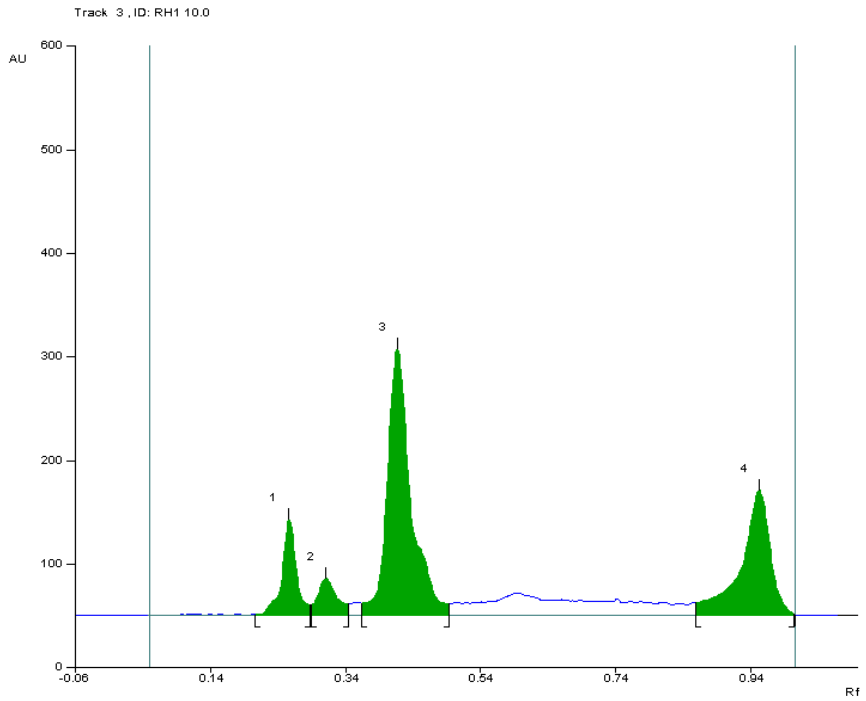
HPTLC Fingerprint - RH1

Sample Preparation

100 mg of extract was weighed and dissolved in 70% methanol to get a concentration of 10mg/ml concentration this is then used for injection.

Chromatographic Conditions

Stationary Phase	: Silica gel 60 F 254
Mobile Phase	: chloroform: methanol (9:1)
Scanning Wavelength	: 404 nm
Applied volume	: 10µl
Development mode	: Ascending mode



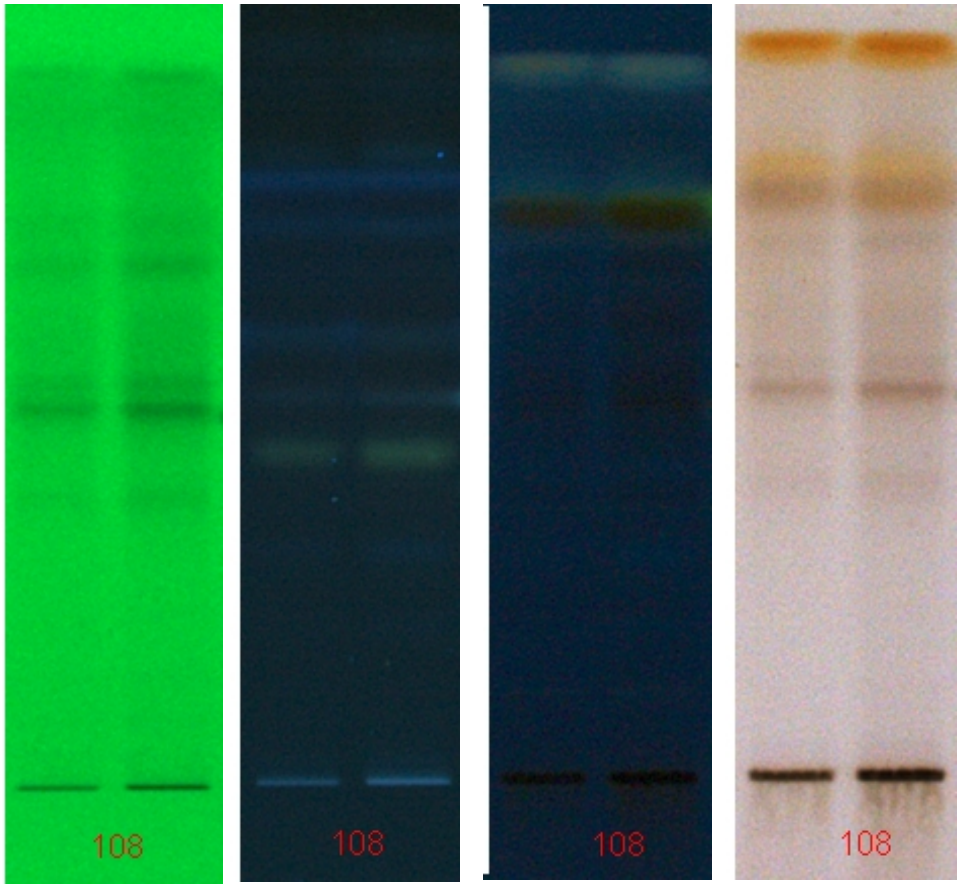
Inference

HPTLC fingerprint of RH -1 shows four peaks at Rf values 0.25, 0.31, 0.41 & 0.95. The peak correspond to the Rf value 0.31 has maximum peak area of 7256.5. At this stage it is difficult to confirm the individual components present in the extract, but from SRM lab experience on phytochemical analysis, They suggest that the major peaks found in the fingerprint may be acidic glycosides / resins. Since, in the present chromatographic conditions, the above mentioned components will be eluted easy.

CHROMATOGRAPHIC CONDITION FOR HPTLC FINGER PRINT

HPTLC WAS DONE AT SRI RAMACHANDRA UNIVERSITY.

Sample Name	: Chithiramoola Rasayanam
Sample-ID	: 108
Stationary phase	: Silica gel F 254
Mobile phase	: n-Hexane: Ethyl acetate:Formic acid 60:40:2.5 ml)
Scanning wavelength	: 254,298,489 nm
Sample concentration	: 20 mg/ml
Injecting volume	: 5, 10 µl
Development mode	: ascending mode



BIO -CHEMICAL ANALYSIS OF CHITHIRAMOOLA RASAYANAM

The biochemical analysis of the Chithiramoola Rasayanam was carried out in the Biochemistry lab, National Institute Of Siddha.

S.No	EXPERIMENT	OBSERVATION	INFERENCE
1.	Appearance of sample	Light Brown in colour.	
2.	**Solubility:** a. A little(500mg) of the sample was shaken well with distilled water. b. A little(500mg) of the sample was shaken well with con. HCl/Con. H_2So_4	Sparingly soluble	Absence of Silicate
3.	**Action of Heat:** A small amount(500mg) of the sample was taken in a dry test tube and heated gartly at first and then strong.	No white fumes evolved	Absence of Carbonate
4.	**Flame Test:** A small amount(500mg) of the sample was made into a paste with con. HCl in a watch glass and introduced into non-luminous part of the Bunsen flame.	No Bluish green flame appeared.	Absence of Copper
5.	**Ash Test:** A filter paper was soaked into a mixture of sample and dil. cobalt nitrate solution and introduced into the Bunsen flame and ignited.	Yellow colour flame appeared.	Presence of sodium

Preparation of Extract:

5gm of Chithiramoola Rasayanam was weighed accurately and placed in a 250ml clean beaker and added with 50ml of distilled water. Then it was boiled well for about 10 minutes. Then it was cooled and filtered in a 100ml volumetric flask and made up to 100ml with distilled water.

S.No	EXPERIMENT	OBSERVATION	INFERENCE
	I.Test For Acid Radicals		
1.	**Test For Sulphate:** a. 2ml of the above prepared extract was taken in a test tube to this added 2ml of 4% dil ammonium oxalate solution b. 2ml of the above prepared extracts was added with 2ml of dil-HCl was added until the effervescence ceases off. Then 2ml of dil.Barium chloride solution was added.	Cloudy appearance present	Absence of Sulphate
2.	**Test For Chloride:** 2ml of the above prepared extract was added with dil. HCl till the effervescence ceases. Then 2ml of dil.silver nitrate solution was added.	No cloudy appearance.	Absence of Chloride
3.	**Test For Phosphate:** 2ml of the extract was treated with 2ml of dil.ammonium molybdate solution and 2ml of con.HNo3.	Yellow appearance present	Presence of Phosphate
4.	**Test For Carbonate:** 2ml of the extract was treated with 2ml dil. Magnesium sulphate solution	No Cloudy appearance.	Absence of carbonate
5.	**Test For Nitrate:** 1gm of the substance was heated with copper turning and concentrated H2So4 and viewed the test tube vertically down.	No Brown gas evolved.	Absence of Nitrate
6.	**Test For Sulphide:** 1gm of the substance was treated with 2ml of con. HCL	No Rotten Egg Smelling gas.	Absence of Sulphide
7.	**Test For Fluoride & Oxalate:** 2ml of extract was added with 2ml of dil. Acetic acid and 2ml dil.calcium chloride solution and heated.	No Cloudy appearance	Absence of fluoride and oxalate

8.	**Test For Nitrite:** 3drops of the extract was placed on a filter paper, on that-2 drops of dil.acetic acid and 2 drops of dil.Benzidine solution was placed.	No Characteristic changes	Absence of Nitrite
9.	**Test For Borate:** 2 Pinches(50mg) of the substance was made into paste by using dil.sulphuric acid and alcohol (95%) and introduced into the blue flame.	No Bluish green colour flame.	Absence of borate
	II. Test For Basic Radicals		
1.	**Test For Lead:** 2ml of the extract was added with 2ml of dil.potassium iodine solution.	No yellow Precipitate obtained.	Absence of Lead
2.	**Test For Copper:** One pinch(50mg) of substance was made into paste with con. HClin a watch glass and introduced into the non-luminuous part of the flame.	No Blue colour flame No Blue colour precipitate formed.	Absence of copper
3.	**Test For Aluminium:** To the 2ml of extract, dil.sodium hydroxide was added in 5 drops to excess.	Yellow colour appeared.	Presence of aluminium
4.	**Test For Iron:** a. To the 2ml of extract,2ml of dil.ammonium solution was added. b. To the 2ml of extract 2ml thiocyanate solution and 2ml of con HNo3 was added	blood red colour appeared.	Presence of Iron
5.	**Test For Zinc:** To 2ml of the extract, dil.sodium hydroxide solution was added in 5 drops to excess and dil.ammonium chloride was added.	No White precipitate was formed	Absence of Zinc
6.	**Test For Calcium:** 2ml of the extract was added with 2ml of 4% dil.ammonium oxalate solution	No Cloudy appearance and white precipitate was obtained	Absence of calcium
7.	**Test For Magnesium:** To 2ml of extract dil.sodium hydroxide solution was added in drops to excess.	White precipitate was obtained	Absence of Magnesium

8.	**Test For Ammonium:** To 2ml of extract 1 ml of Nessler's reagent and excess of dil.sodium hydroxide solution are added.	No Brown colour appeared	Absence of ammonium
9.	**Test For Potassium:** A pinch(25mg) of substance was treated of with 2ml of dil.sodium nitrite solution and then treated with 2ml of dil.cobalt nitrate in 30% dil.glacial acetic acid.	No Yellowish precipitate was obtained.	Absence of Potassium
10.	**Test For Sodium**: 2 pinches(50mg) of the substance was made into paste by using HCl and introduced into the blue flame of Bunsen burner.	yellow colour flame appeared	Presence of sodium
11.	**Test For Mercury**: 2ml of the extract was treated with 2ml of dil.sodium hydroxide solution.	No yellow precipitate was obtained	Absence of mercury
12.	**Test For Arsenic:** 2ml of the extract was treated with 2ml of dil.sodium hydroxide solution.	No brownwish red precipitate was obtained	Absence of arsenic
	III. Miscellaneous		
1.	**Test For Starch:** 2ml of extract was treated with weak dil.iodine solution	No blue colour developed	Absence of starch
2.	**Test For Reducing Sugar:** 5ml of Benedict's qualitative solution was taken in a test tube and allowed to boil for 2 minutes and added 8 to 10 drops of the extract and again boil it for 2 minutes. The colour changes are noted.	Brick red colour not developed	Presence of reducing sugar
3.	**Test For The Alkaloids:** a) 2ml of the extract was treated with 2ml of dil.potassium iodide solution. b) 2ml of the extract was treated with 2ml of dil.picric acid. c) 2ml of the extract was treated with 2ml of dil.phosphotungstic acid.	. Yellow colour developed	Presence of Alkaloid
4.	**Test For Tannic Acid:** 2ml of extract was treated with 2ml of dil.ferric chloride solution	No black precipitate was obtained	Presence of Tannic acid

5.	**Test For Unsaturated Compound:** To the 2ml of extract 2ml of dil.Potassium permanganate solution was added.	Potassium permanganate was not decolourised	Absence of unsaturated compound
6.	**Test For Amino Acid:** 2 drops of the extract was placed on a filter paper and dried well. 20ml of Biurette reagent was added.	violet colour developed	Presence of amino acids
7.	**Test For Type Of Compound:** 2ml of the extract was treated with 2 ml of dil.ferric chloride solution.	No green colour developed No red colour developed No violet colour developed No blue colour developed	Absence of oxyquinole pinephrine and pyro catechol Anti pyrine, Aliphatic amino acids and meconic acid are absent Apomorphine salicylate and Resorcinol are absent Morphine, Phenol cresol and hydro uinone are absent

TOXICITY STUDY

ACUTE AND SUB ACUTE TOXICITY STUDY ON CHITHIRAMOOLA RASAYANAM IN RODENTS

Animals:

Mice of either sex weighing 25-30g and rats weighing 210-240g were obtained from the animal house of Vels University. The animals were used with the approval of the Institute Animal Ethics Committee and obtained from Vels University, Chennai. They were fed with a balanced standard pellet diet and maintained under standard laboratory conditions, providing 24-28^{0}C temperature, standard light cycle (12 h light, 12 h dark) and water ad libitum. Animals were kept in cages with raised floors of wide mesh to prevent coprophagy. Animal welfare guidelines were observed during the maintenance period and experimentation. The rats were randomly assigned to control and different treatment groups, six animals per group. (Approval number: XIII/VELS/PCOL/32/2000/CPCSEA/IAEC/08.08.2012). The animals were acclimatized for one week under laboratory conditions.

ACUTE TOXICITY STUDY-OECD 425 GUIDELINES

Acute oral toxicity test for the Chithiramoola Rasayanam was carried out as per OECD Guidelines 425. As with other sequential test designs, care was taken to ensure that animals are available in the appropriate size and age range for the entire study. The test substance is administered in a single dose by gavage using a stomach tube or a suitable intubation cannula. The fasted body weight of each animal is determined and the dose is calculated according to the body weight. After the substance has been administered, food was withheld for a further 2 hours in mice. The animals were observed continuously for the first 4 h and then each hour for the next 24 h and at 6 hourly intervals for the following 48 h after administering of the test drug, to observe any death or changes in general behaviour and other physiological activities. Single animals are dosed in sequence usually at 48 h intervals. However, the time interval between dosing is determined by the onset, duration, and severity of toxic signs. Treatment of an animal at the next dose was delayed until one is confident of survival of the previously dosed animal.

Observation of toxicity signs: General behavior, respiratory pattern, cardiovascular signs, motor activities, reflexes, change in skin and fur, mortality and the body weight changes were monitored daily. The time of onset, intensity, and duration of these signs, if any, were recorded.

Stock solution and Acute toxicity study

Acute oral toxicity study was performed as per OECD-425 guidelines. Mice (n=6) of either sex selected by random sampling technique were used for acute toxicity study. The animals were kept fasting for overnight providing only water, after which the Chithiramoola Rasayanam in 2% CMC was administered orally at the different dose levels in up and down dosing schedule according to body weight by gastric intubation and observed for 14 days.

SUB-ACUTE TOXICITY

In a 28-days sub acute toxicity study, twenty four rats of either sex were divided into four groups of 6 rats each. Groups of rats I, II and III were administered daily with the Chithiramoola Rasayanam (p.o.) for 28 days at a dose of 100, 250 and 500mg/kg respectively. The animals were then observed daily for gross behavioural changes and any other signs of sub-acute toxicity. The weight of each rat was recorded on day 0 and weekly throughout the course of the study, food and water consumption per rat was calculated. At the end of the 28 days they were fasted overnight, each animal was anaesthetized with diethylether, following which they were then dissected and blood samples were obtained by retro orbital puncture into heparinised tubes. The blood sample collected from each rat was centrifuged with 3000 X g at 4^{o}C for 10 min to separate the serum and used for the biochemical assays.

Hematological and blood biochemical analyses:

At the end of the study, all animals were kept fasted for 16-18 h and then anesthetized with anesthetic ether on the 28th day. Blood samples for hematological and blood chemical analyses were taken from retro orbital vein. Heparinized blood samples were taken for determining complete blood count (white blood cell count, differential white blood cell count, platelet count, red blood cell count, and hemoglobin etc..) by semiautomated hematology analyzer. The serum from non-heparinized blood was carefully collected for blood chemistry and enzyme analysis (Glucose, creatinine, total

protein, albumin, total and direct bilirubins, serum glutamate-oxaloacetate transaminase (SGOT), serum glutamate pyruvate transaminase (SGPT), and alkaline phosphatase (ALP) etc..) were automatically determined using autoanalyzer.

Necropsy:

All rats were sacrificed after the blood collection. The positions, shapes, sizes and colors of internal organs were evaluated. The Spleen, Testes, Pancreas, Lung, Liver, Brain, Heart, Stomach, Intestine, Bone, Ovary, and Kidney tissues were excised from all rats to visually detect gross lesions, and weighed to determine relative organs, weights and preserved in 10% neutral formalin for histopathological assessment. The tissues were embedded in paraffin, and then sectioned, stained with haematoxylin and eosin and were examined microscopically.

Statistical analysis

Values were represented as mean ± SEM. Data were analysed using one-way analysis of variance (ANOVA) and group means were compared using the Dunnet's 't' Test using GraphPad Instat-V3 software. P values < 0.05 were considered significant.

RESULTS AND DISCUSSION

The acute toxicity study of the Chithiramoola Rasayanam indicated no changes in the behaviour and in the sensory nervous system responses in the animals. Also no adverse gastrointestinal effects were observed in the male and female mice used in the experiment. All the mice that received up to 5.0g/kg dose of the Chithiramoola Rasayanam survived beyond the 24 hours of observation. Hence the dose was fixed as 100, 250 and 500mg/kg for further sub acute toxicity study.

During the sub-acute toxicity tests, the results obtained on the average daily water, food intake and periodical weight gain ($P<0.01$). The eating and drinking habit and behavior of all the animals used were normal in both vehicle-treated and Chithiramoola Rasayanam treated animals. The results revealed that essential organs such as the liver, kidney, spleen and testes were not adversely affected during the sub-acute administration. Acute and sub-acute oral administration of Chithiramoola Rasayanam did not cause any significant changes in gross behavioural effects in rodents.

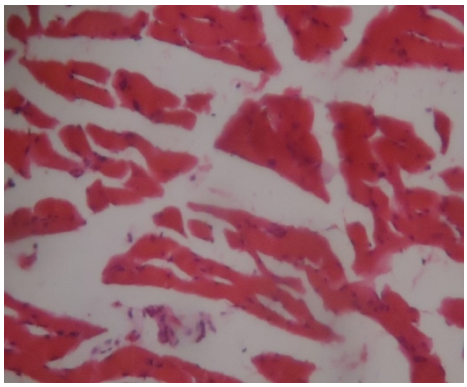
The changes in the SGPT, total protein and albumin concentrations following the administration of 0.25,0.5 g/kg of Chithiramoola Rasayanam was observed ($P<0.01$) and showed significant alterations in the serum HDL, LDL and Uric acid ($P<0.01$) concentration when compared to the control. On the other hand, there was a significant increase in the triglyceride concentration but statistically not significant. Significant alterations were observed in monocyte count. No other heamatological changes were identified. Macroscopically, the liver, spleen, lung, testis and the kidney showed no discolouration and the textures were consistent when compared with the control groups. Slightly significant reduction in weight was observed in the vital organs like Spleen, brain and kidney. Histopathological examination revealed that the spleens, livers, lung, testes and the kidneys of rats administered with Chithiramoola Rasayanam showed no differences relative to those of the control group at the two dose levels, though there was focal proximal tubular epithelial necrosis in the kidney at 5.0 g/kg. These results indicate that Chithiramoola Rasayanam at 0.5 g/kg body weight is not toxic to the liver, spleen and testes of rat but has a minor effect on the lungs and kidney. Thus, since there was no any significant changes in serum levels of glucose, triglycerides concentration following a 28 days treatment of the Chithiramoola Rasayanam, it may indicate therefore, that it is not toxic to the animal.

In conclusion, the present results show that Chithiramoola Rasayanam possesses negligible toxicity as indicated in our rat model. No deaths or signs of toxicity were observed in the rats that received the Chithiramoola Rasayanam up to an oral acute dose of 5g/kg thus establishing its safety in use.

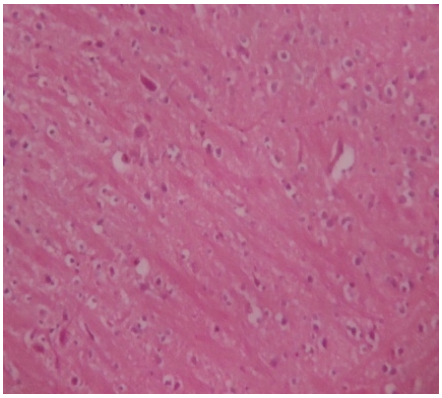
HISTOPATHOLOGY

Chithiramoola Rasayanam(500mg)

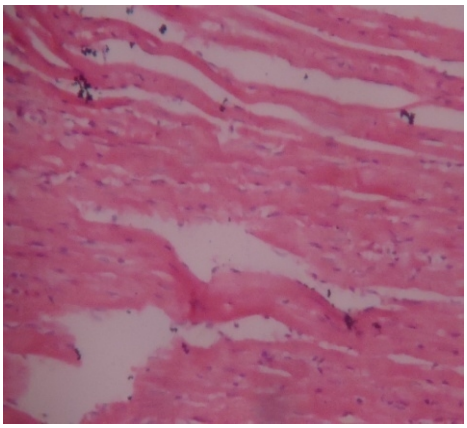
Bone



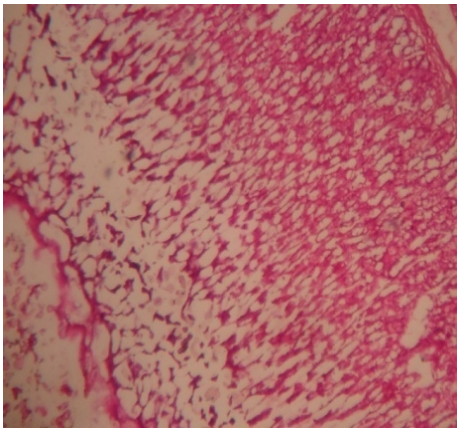
Brain



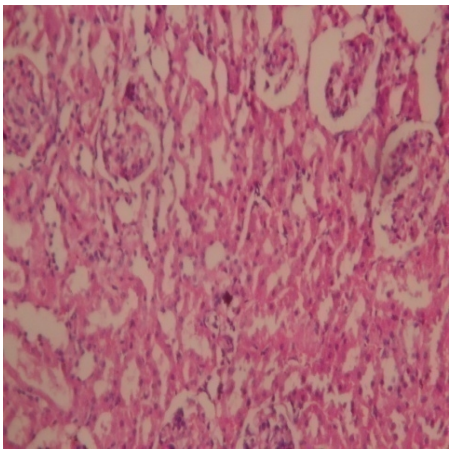
Heart



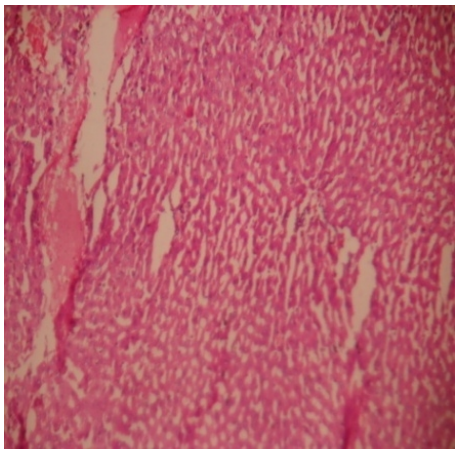
Intestine



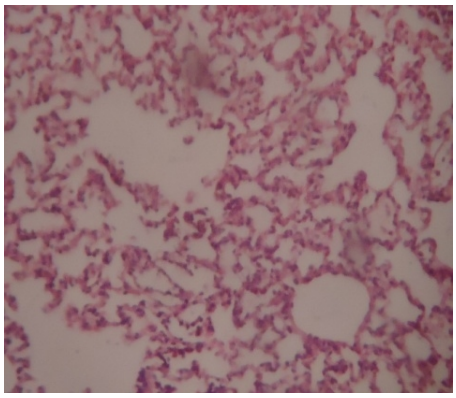
Kidney



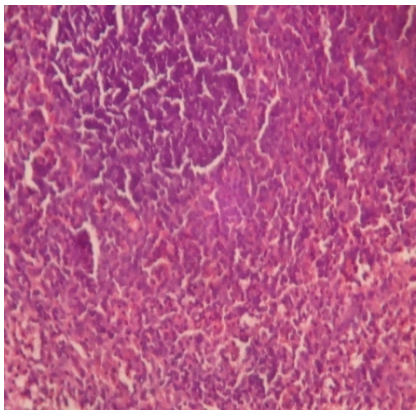
Liver



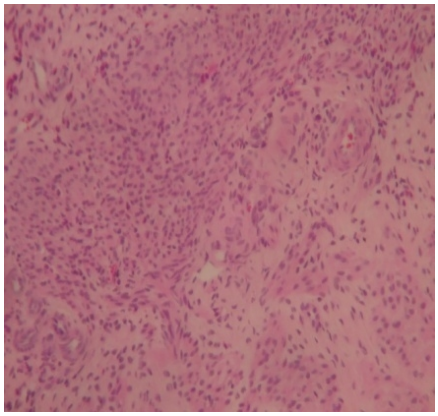
Lung



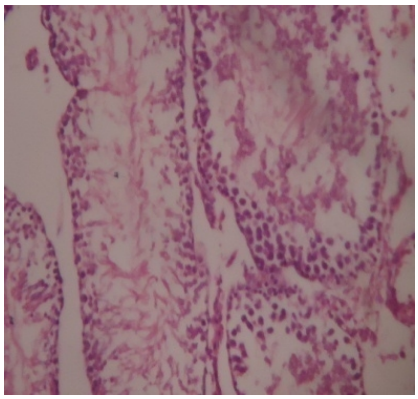
Spleen



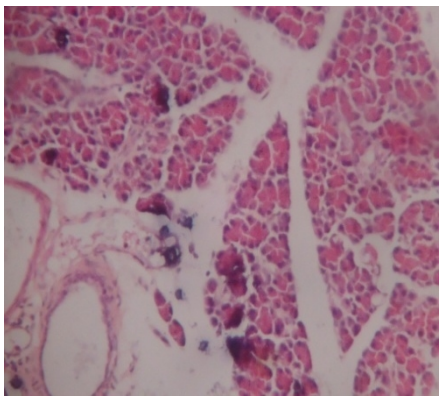
Ovary



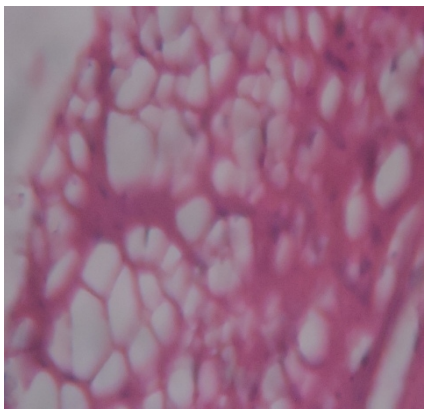
Testis



Pancreas



Stomach



PHARMACOLOGICAL STUDY

ACUTE ANTI INFLAMMATORY ACTIVITY OF CHITHIRAMOOLA RASAYANAM IN RATS

MATERIALS AND METHODS

Animals

Male albino Wistar rats (190-210 g) were obtained from the animal house of the School of Pharmaceutical Sciences, Vels University, Chennai. They were kept at standard environmental conditions (12/12-h light/dark cycle) and were allowed free access to food and water. Before each test, the animals were fasted for 24 h with free access to water. The rats were randomly divided into test and control groups, each group consisted of age and weight matched rats (n =6). The experimental protocol was approved by the animal ethical committee of Vels University.

(Approval number:XIII/VELS/PCOL/32/2000/CPCSEA/IAEC/08.08.2012)

ACUTE ANTI-INFLAMMATORY EVALUATION

Formalin induced method

Male Albino Wistar rats, 190–210 g, were kept in Polypropylene cages with free access to food and water. Testing took place in the middle of the light period of a 12:12-h light:dark cycle. The Chithiramoola Rasayanam was suspended in vehicle (2% carboxy methyl cellulose (CMC) in saline) and administered orally at a dose of 100, 250, 500 mg/kg and 45mg/kg for Diclofenac sodium. The rats were divided in to five groups (n=6) and the first group treated with saline (5ml/kg), second group treated with Diclofenac sodium (45mg/kg) and third, fourth and fifth group treated with Chithiramoola Rasayanam 100, 250 and 500mg/kg respectively through orally. Oedema was produced by subplantar injection of formalin in the right hindpaw of each rat one hour after the administration of corresponding drugs. The paw volume was measured at 1,2,3 and 4 hr after the injection of formalin using the plethysmometer. Mean increase in the paw volume of oedema was measured.

Statistical analysis

The data are expressed as mean±SEM. Student t-test followed by Dunnet 't' test was used to determine significant differences between groups. *p*-values less than 0.05 were considered as indicative of significance.

RESULT AND DISCUSSION

In Indian system of medicine, certain drugs are claimed to provide relief of inflammation. The claimed therapeutic reputation has to be verified in a scientific manner. Inflammation is a complex process and ROS play an important role in the pathogenesis of inflammatory diseases. The main action of anti-inflammatory agents in the inhibition of cyclooxygenase enzyme, which are responsible for conversion of arachidonic acid to prostaglandin (PG). The extracellular activity of these enzymes is said to be related to acute and chronic inflammation. NSAID'S act either by inhibiting these lysosomal enzymes (Cyclooxygenase). In the Present Study, the result suggests that the test drug Chitramoola Rasayanam becomes significant within two hour, during the phagocytic phase of formalin-induced inflammation. Inflammation is a normal protective response to tissue injury caused by physical trauma, noxious chemicals or microbiologic agents. Inflammation is body's response to inactivate or destroy the invading organisms, remove irritants and set stage for tissue repair. Inflammation is triggered by the release of chemical mediators from the injured tissues and migrating cells. The specific chemical mediators vary with the type of inflammatory process and include amines such as histamine, serotonin, lipids such as prostaglandins and small peptides such as Kinins. Prostaglandins (PGs) play significant role in different phases of inflammatory reactions. PGs elicit pain by direct stimulation of sensory nerve endings and also sensitize sensory nerve endings to other pain provoking stimuli. Moreover, PGs especially PGE**1** was reported to act on cell membrane during inflammatory conditions leading to changes in lipoprotein structure of cell membrane. This causes destabilization of cell membrane furthering to degenerative cellular changes.

An important feature of the formalin test in rodents is that the animal show two phases of nociceptive behavior which possibly involves two distinctly different stimuli. The first phase starts immediately after injection of the formalin and lasts for 3-5 min. Evidences show that effect on the opioid receptors is one of the main ways involved in exertion of antinociceptive effects in this phase. The most widely used primary test to screen new anti-inflammatory agents measures the ability of a compound to reduce local

edema induced in the rat paw by injection of an irritant agent. This edema depends on the participation of kinins and polymorphonuclear leukocytes with their proinflammatory factors including prostaglandins.

The development of edema in the paw of the rat after the injection of irritants has been described as a biphasic event. The initial phase, observed around 1 h, is attributed to the release of histamine and serotonin; the second, accelerating phase of swelling is due to the release of prostaglandin-like substances. It has been reported that the second phase of edema is sensitive to both clinically useful steroidal and nonsteroidal anti-inflammatory agents.

The specific chemical mediators vary with the type of inflammatory process and include amines such as histamine, serotonin, lipids such as prostaglandins and small peptides such as Kinins. Formalin-induced pain is caused primarily by peripheral tissue inflammation. Acute inflammation may last for relatively shorter duration, ranging from few minutes to few days. Exudation of fluid and plasma proteins, emigration of leukocytes, and predominantly neutrophils, are characteristic changes. The Chithiramoola Rasayanam as well as diclofenac showed Anti-phleogestic activity. It can be assumed that the test drug Chithiramoola Rasayanam exert its anti-inflammatory effect through mechanism similar NSAIDs. Formalin-induced paw oedema is one of the most suitable test procedures to screen chronic anti-inflammatory agents, as it closely resembled human arthritis. The nociceptive effect of formalin is also biphasic; an early neurogenic component followed by a later tissue-mediated response. The result suggests the usefulness of Chithiramoola Rasayanam in the treatment of inflammation associated diseases like arthritis. The pattern of anti-inflammatory activity exhibited by this Chithiramoola Rasayanam was similar to that of diclofenac which suggests that the activity may be mediated by cyclooxygenase I and II inhibition. This anti-inflammatory activity was found to be statistically significant ($P<0.01$) at all the concentration used after 120minutes of drug treatment.

ANALGESIC ACTIVITY OF CHITHIRAMOOLA RASAYANAM IN MICE

Mice of either sex with an intial weight 21-41g of mice were obtained from the animal house of the School of Pharmaceutical Sciences, Vels University, Chennai. They were kept at standard environmental conditions (12/12-h light/dark cycle) and were allowed free access to food and water. Before each test, the animals were fasted for 24 h with free access to water. The rats were randomly divided into test and control groups, each group consisted of age and weight matched rats (n =6). The experimental protocol was approved by the animal ethical committee of Vels University.
(Approval number: XIII/VELS/PCOL/32/2000/CPCSEA/IAEC/08.08.2012)

PROCEDURE

The present study was undertaken to study the analgesic activity of Chithiramoola Rasayanam- A Siddha Drug and was assessed by the Eddy's hot plate method in mice. The following procedure has been used in this study. Groups of 6 mice of either sex with an initial weight of 22 to 41 g were used. The hot plate, which is commercially available, consists of a electrically heated surface. The temperature is controlled for 55° to 56 °C. The animals are placed on the hot plate and the time until either licking or jumping occurs is recorded by a stop-watch. The latency is recorded before and after 15, 30, 45, 60 and 90 min following oral or intra peritoneal administration of the standard or the Chithiramoola Rasayanam.

RESULTS AND DISCUSSION

The prolongation of the latency times comparing the values before and after administration of the Chithiramoola Rasayanam with the experimental groups used for statistical comparison using the One way-ANOVA followed by Dunnet test. Alternatively, the values which exceed the value before administration for 50% or 100% can be regarded as positive. Doses of pentazocine hydrochloride (5mg/kg) and 100, 250 and 500mg/kg p.o.. Chithiramoola Rasayanam was considered for treatment. Analgesic effect lasted for a period of 90min was found to possess significant ($P<0.01$) analgesic activity at 250 and 500mg/kg dose levels. There was increase in % of reaction time by dose dependant manner.

DISEASE ASPECT

SIDDHA ASPECT

சூலைநோய்[6]

வேறுபெயர்கள் :

முருக்கல்நோய், குத்தல் நோய், குடல்வலி எனப் பல பெயர்களுண்டு.

இயல்பு :

சூலம் என்னும் கருவியால் குத்துங்கால் உண்டாகும் வலியை யொத்த நோய்க்கு சூலை நோய் எனப் பெயராயிற்று. சூலம் என்ற கருவியால் குத்துங்காலுண்டாகும் வலியையொத்த நோயை, இரைப்பை, குடல் பக்கம் (விலா), மூட்டுகள் இவற்றிலுண்டாகும் குத்தல் நோயாம்.

நோய்வரும் வழி :

“சார்வான சூலைவரு மாறு கேளாய்-
தக்கசிறைப் பட்டிருக்குந் தீமையாலும்
ஆர்வான வறச்சுடுசோ றருந்த லாலும்
அறவுமே சலிப்பாலு மோட லாலுந்
தார்வான சபைமிகுந்த சண்டை யாலும்
தகையான துவர்ப்பொசிப்பு புகைத்த லாலும்
வேர்வான மோகத்தின் புணர்ச்சி யாலும்
மிகுந்தபசி யுறுதலினாற் சூலை யாமே”

“ஆமென்ற வன்னத்துக் கிறுதி பண்ணி
யதிகபர தேசிகளை யடித்த பேர்க்குங்
காமென்ற கற்புடைய மங்கை மாரைக்
கருதியே மனத்துளிச் சித்த பேர்க்கும்
வாமென்ற வாழ்மரத்தை வெட்டி னோர்க்கும்
வழிமறித்து பொருள்பறித்த மதிகே டர்க்கும்
ஏமென்ற எச்சிறனைக் கவர்ந்த பேர்க்கும்
இகத்திலே நோவெய்திச் சூலை யாமே”

பொருள் :

சிறைப்பட்டிருத்தல், மிகச் சூடான பண்டங்களை உண்ணல், மனச்சலிப்பு கொள்ளல், ஓடல், தன் வன்மைக்கு மிகுந்த சண்டையிடல், துவர்ப்புப் பொருட்களை அடிக்கடிக்கு கொள்ளல், புகை பிடித்தல், அளவிற்கு விஞ்சிப் பெண்கலவி செய்தல் ஆகியவற்றால் சூலை நோய் உண்டாகும்.

ஏழைகட்கு உணவிடாமல் அவர்களை அடித்தோட்டியவருக்கும், மிகுந்த பெண்ணாசையால் கற்புடைய மங்கையரை விரும்பியவர்க்கும், பயன்தரக் கூடிய மரங்களை வெட்டி வீழ்த்தியவருக்கும், பொருளிச்சையால் வழிபறிக்கும் அறிவிலிகளுக்கும், தன் எச்சிலைத் தெரியாமல் பிறர்க்கு கொடுப்பவர்க்கும் இந்நோய் உண்டாகும் என நூற்கள் கூறும்.

முற்குறிகள் :

உடல் குளிர்ந்தாற் போற்றோன்றி, மூக்கில் நீர் பாய்தல், கண் சிவத்தல், சற்று சுரம் காய்தல், உடல் வனப்புக் குறைதல் என்னும் முற் குறிகளையும் காட்டி, பக்கம், கைகால், பூட்டுகள் இவைகளில் குத்தலை உண்டாக்கும்.

சூலை குறிகுணம் :

- நோய் பிறக்கும் இடங்களில் சற்று வீங்கிச் சிவந்து, குத்தலை உண்டாக்கும்
- குத்தலின் வன்மைகேற்ப, மிகுசுரம், வாந்தி, மயக்கம், தலைநோய், கண் சிவத்தல், காதுகேளாமை, அறிவு குன்றல், வாய் பிதற்றல் ஏற்படும்.
- நோயின் வலி தாங்க முடியாது உடல் வியர்த்தல், கையும் காலும் சில்லிட்டு போதல், மூக்கு நுனி நீண்டு போதல், கண் பஞ்சடைதல் ஏற்படும்.

குற்ற முதலிய வேறுபாடுகள் :

"நெடுவாத சார்வதுமின்றிச் சூலை வாராது"

வளிக்குற்றம் மிகுந்து உடலிற்றங்கி அதனால் கால்களில் (வாயுகளில்) மேல் நோக்குக் கால், கீழ்நோக்குக் கால்களின் வன்மைமிகுந்து நோயைப் பிறப்பிக்கும்.

நோயெழுந்த காலை இரு குற்றங்களுள் ஒன்றேனும், இரண்டேனும், கூடுமாதலின், நோய் வன்மையும் பெருகும்.

உடலின் உறுப்புகளில் குத்தல், குடைதல், வலித்தல், புரட்டல் என்னுங் குறிகளையும் சிறுநீர், எரு இவற்றை வெளியேறச் செய்யாது கட்டுதலாகிய செயலையும் உண்டாக்கும்.

OSTEOARTHRITIS[34]

Osteoarthritis is also erroneously called degenerative joint disease, represent diarthroidal (movable, synovial-lined) joint. Osteoarthritis (OA) is characterized by focal loss of articular cartilage and proliferation and remodelling of bones around the joint to form osteophyte . Inflammation can be a feature of osteoarthritis.

Osteoarthritis represents an imbalance in destruction and synthetic process of the cartilage lead to the erosion, decreased concentration and viscosity of the synovial fluid ,decreased lubricating and cushioning properties . There is also an underlying inflammation of the synovium as well as damage in the subchondral bone.

EPIDEMIOLOGY:

- Osteoarthritis (OA) affects 10-15% of world population.[30]
- About 5.7% of population of India has Osteoarthritis (OA). [31]
- In general prevalence of osteoarthritis increases with age
- 80% of people affected by 40 years. More than 50% have bilateral osteoarthritis
- Women have great tendency than men.[32]
- Genetic tendency in knee osteoarthritis is twice as osteoarthritis hip
- Knee osteoarthritis is the leading cause of chronic disability in developed countries among elders, some people are unable to walk independly from bed to bathroom because of knee osteoarthritis.

RISK FACTORS[21]:

Age	: Repetitive stress, e.g. vocations, obesity
Female	: obesity
Genetic factors	: Prior inflammatory joint disease
Major joint trauma	: Metabolic / Endocrine disorder

TYPES:

Primary / Idiopathic osteoarthritis

Secondary osteoarthritis

Primary /idiopathic osteoarthritis.

Localised Osteoarthritis :

Hands (Herbenden's node,bouchard,s node)

Feet (Hallux valgus,Hallux rigidus)

Knee:

Medial Compartment knee osteoarthritis

Lateral Compartment knee osteoarthritis

Patellofemoral Compartment knee osteoarthritis

Hip:

Eccentric (superior)

Concentric (axial, medial)

Diffuse (coxae senilis)

Secondary Osteoarthritis.

Trauma. e.g. sports

Congenital or developmental:e.g. valgus, varus deformity.

Metabolic, e.g. Wilsons disease

Endocrine, e.g. obesity, diabetes mellitus.

CLINICAL FEATURES:

Pain

Swelling

Stiffness

Restricted movements

Crepitus

Tenderness

COMPLICATIONS:

Joint deformity

Subluxation

Ankylosis

Intra articular loose bodies

STUDY DESIGN :

The study was conducted on patients with Soolai (Osteoarthritis) patients satisfying the inclusion criteria.

Study place : NIS (OPD,IPD)

Period : 12 months

Sample size : 20 patients (both sex)

Weight : 35-85 kg

Dose : 5gm bid

Duration : 40 days

SUBJECT SELECTION:

Inclusion Criteria:

1. Age :35-65 years
2. Sex :both male and female
3. Weight :35-85 kg
4. Patient, having symptoms of

   One or both knee joints pain

   One or both knee joint swelling

   Stiffness

   Crepitus

   Restricted movements of knee joint

   Any of the 2 clinical symptoms.
5. Patienst who are willing to provide blood for lab investigation.
6. patients who are willing to undergo Radiological investigation.
7. Patients willing to sign the informed consent stating that he/she will conscientiously stick to the treatment during 40 days but can opt out of the trial of his/her own conscious discretion.

Exclusion criteria:

Rheumatoid arthritis

Gouty arthritis

Tuberculosis

Viral fever

History of trauma

Any other serious illness

Withdrawal criteria:

1. Development of any adverse reaction
2. Occurrence of any other serious illness
3. Non co-operation of the patient

TESTS AND ASSESMENTS

C. Clinical assessment

Siddha assessment

D. Laboratory Investigations

Routine investigations

Clinical assessment:

Patient, having symptoms of

One or both knee joints pain

One or both knee joint swelling

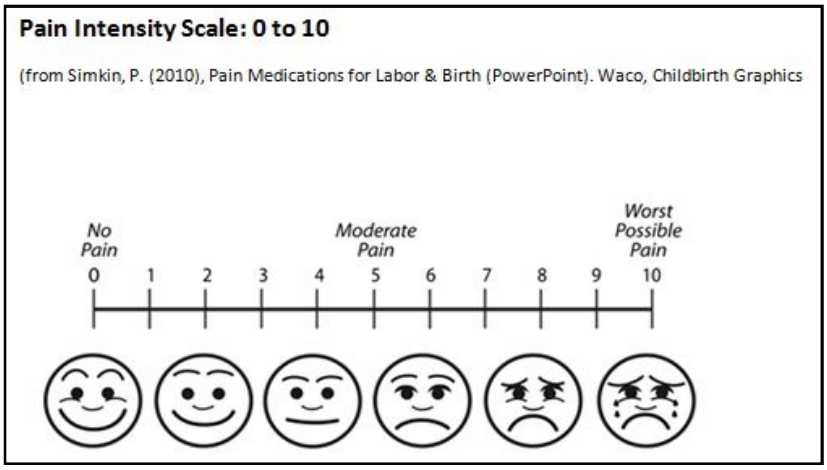
Stiffness

Crepitus

Restricted movements of knee joint

Any of the 2 clinical symptoms.

PAIN SCALE



Grade 0 : No Pain

Grade 1 -3 : Mild pain

Grade 4-6 : Moderate pain

Grade 7-10 : Severe pain

2. RESTRICTED MOVEMENT ASSESSMENT SCALE:

Gradation of movements:

Grade 1 - Fit for all activities, do their work without support.

Grade II - Mild pain present in knee joint, mild restricted movements.

Grade II - Pain present in knee joint, moderate restriction of movements.

Grade IV - Severe pain, bed ridden.

(Reference: Clinical manual for nursing practice (National Institute of Health Warren Grant Magnuson Clinical Centre)

5. Patient who are willing to provide blood for lab investigation.

6. patient who are willing to undergo Radiological investigation.

7. Patient willing to sign the informed consent stating that he/she will conscientiously stick to the treatment during 40 days but can opt out of the trial of his/her own conscious discretion.

INVESTIGATION/ASSESSMENT:

SIDDHA PARAMETERS:

Enn vagai thervu**:**

1.Naa

2.Niram

3.Mozhi

4.Vizhi

5.Sparisam

6.Naadi

7.Malam

8.Moothiram

- Neerkuri
- Neikuri

ROUTINE INVESTIGATION

Blood

Hb (gm/dl)

Total WBC Count(Cells/cumm)

DC- Polymorphs (%)

Lymphocytes (%)

Eosinophils (%)

Monocytes (%)

Basophils (%)

Total RBC count (Million cells / cu.mm)

ESR (mm/hr)

Blood glucose(mg/dl): (Fasting)

(Post – prandial)

Blood urea

Serum creatinine

Liver function test:

SGOT

SGPT

Serum total bilirubin

Serum bilirubin (direct)

Serum bilirubin(indirect)

Serum alkaline phosphatase

Serum total protein

Serum albumin

Serum globulin

Serum calcium

Serum phosphorus

Serum uric acid

Lipid Profile

Serum cholesterol(mg/dl)

HDL cholesterol(mg/dl)

LDL cholesterol(mg/dl)

VLDL cholesterol(mg/dl)

Serum triglycerides (mg/dl)

X-RAY: Knee joint

VDRL:

Urine examination:

Albumin

Sugar (fasting and post prandial)

Deposits

Specific investigation:

ASO Titre

RA Factor

CRP

X-RAY

STUDY ENROLLMENT:

- In this pilot study, patients reporting at the OPD with the clinical symptoms of pain in one or both knee joints, swelling, crepitus, restricted movements, stiffness will be examined clinically for enrolling in the study based on the inclusion and exclusion criteria.

- The patients who are to be enrolled would be informed (Form IV C) about the study, trial drug, possible outcomes and the objectives of the study in the language and terms understandable to them.
- After ascertaining the patient's willingness, informed consent would be obtained in writing from them in the consent form (Form IV-A).
- All these patients will be given unique registration card in which patients' Registration number of the study, Address, Phone number and Doctors phone number etc. will be given, so as to report easily should any complications arise.
- Complete clinical history, complaints and duration, examination findings-- all would be recorded symptom in the prescribed Proforma in the history and clinical assessment forms separately. Screening Form- I will be filled up; Form I-A, Form –II and Form –III will be used for recording the patients' history, clinical examination of symptoms and signs and laboratory investigations respectively.
- Patients would be advised to take the trial drug and appropriate dietary advice (Form IV-D) would be given according to the patient's perfect understanding.

CONDUCT OF THE STUDY:

- Osteoarthritis patient who satisfying the inclusion criteria will be admitted to the trial.
- Patient informed consent will be obtained
- For OP patients ,they should visit the hospital once in 7 days. At each clinical visit clinical assessment is done and prognosis is noted.
- For IP patients clinical assessment is daily and prognosis is noted
- Laboratory investigations are done before the trial started and at end of the trial for both OP & IP patients .
- For IP patients, who is not in a situation to stay in the hospital for a long time is advised to attend the OPD for the continuation of the treatment. After the end of the treatment also, the patient is advised to visit the OPD for another 2months for follow-up. If any trial patient who fails to collect the trial drug on the prescribed day but wants to continue in the trial from the next day or two, he/ she will be allowed, but defaulters of one week and more will not be allowed to continue and be withdrawn from the study with fresh case being included.

DATA MANAGEMENT

- After enrolling the patient in the study, a separate file for each patient will be opened and all forms will be filed in the file. Study No. and Patient No. will be entered on the top of file for easy identification. Whenever study patient visits OPD during the study period, the respective patient file will be taken and necessary recordings will be made at the assessment form or other suitable form.
- The screening forms will be filed separately.
- The Data recordings will be monitored for completion by HOD and adverse reactions by Pharmacovigilance department of NIS . All forms will be further scrutinized in presence of Investigators by Sr. Research Officer (Statistics) for logical errors and incompleteness of data to avoid any bias. No modification in the results is permitted for unbiased reports.

OUTCOME:

Primary outcome is mainly assessed by

- Reduction of clinical symptoms.

DISCUSSION

The principle aim of this study was to assess the pre-clinical safety and efficacy and to evaluate the therapeutic efficacy of the drug *Chithiramoola Rasayanam* in the management of Soolai.

As per Siddha text, in Soolai Vathaa humors were deranged. Vatha thathu is responsible for the functioning of the Udal thathukal uniformly. Hence derangement of the Vatha kutram leads to impairment in Udal thathukal and in turn produces symptoms like tiredness, body ache, difficult to flexion and extention of joint, pain in all joint ext.
The literary evidence from the text Pulipani Vaithiyam 500 strongly supports the Anti-inflammatory activity and Analgesic activity.The trial drug Chithiramoola Rasayanam was effective in the management of Soolai.

Biochemical analysis:

Biochemical analysis of the drug Chithiramoola Rasayanam reveals the presence of sodium, phosphate, iron, tannic acid, Aluminium, phosphate and alkaloids.

Sodium[25]:

Important for acid-base balance.

50% of body Sodium present in bone.*

Required for normal muscle irritability and cell permeability.

Sodium deficiency causes muscle cramps.

Sodium reduces prostaglandin synthesis.

Iron:

Iron prevents lipid peroxidation and protect the cell against the free radicals including superoxide.

Peroxidase, the lysosomal enzyme, is required for electron transport chain and oxidative phagocytosis and killing of bacteria by neutrophils.

It is a component of heam which is required for the formation of hemoglobin .

Phosphate:

It is essential for the development of bones and teeth.*

Toxicological studies:

The acute toxicity study of the Chithiramoola Rasayanam indicated no changes in the behaviour and in the sensory nervous system responses in the animals. Also no adverse gastrointestinal effects were observed in the male and female mice used in the experiment. All the mice that received up to 5.0g/kg dose of the Chithiramoola Rasayanam survived beyond the 24 hours of observation

Toxicological study results indicate that Chithiramoola Rasayanam at0.25, 0.5 g/kg body weight is not toxic to the liver, spleen and testes of rat but has a minor effect on the lungs and kidney. Thus, since there was no any significant changes in serum levels of glucose, triglycerides concentration following a 28 days treatment of the Chithiramoola Rasayanam, it may indicate therefore, that it is not toxic to the animal.

In conclusion, the present results show that Chithiramoola Rasayanam possesses negligible toxicity as indicated in our rat model. No deaths or signs of toxicity were observed in the rats that received the Chithiramoola Rasayanam up to an oral acute dose of 5g/kg thus establishing its safety in use.

Pharmacological Studies

Pharmacological Studies suggests that the test drug Chithiramoola Rasayanam becomes significant within two hour, during the phagocytic phase of formalin-induced inflammation. The Chithiramoola Rasayanam as well as diclofenac showed Anti-phleogestic activity. The pattern of Anti-inflammatory activity exhibited by this Chithiramoola Rasayanam was similar to that of diclofenac which suggests that the

activity may be mediated by cyclooxygenase I and II inhibition. This anti-inflammatory activity was found to be statistically significant ($P<0.01$) at all the concentration used after 120minutes of drug treatment.

Clinical observation:

From the clinical study 95% of patients relieved from pain,89% of patients relieved from swelling, 71% of patients relieved from stiffness ,50% of patients relieved from crepitus, 85% of patients relieved from restricted movements and no adverse effects were observed during trial period**.**

Bio-statistics:

Statistically, the paired 't' test shows statistical significance for the symptoms before and after the treatment.($p<0.0001$).

SUMMARY

The literary evidence strongly supports the Anti-inflammatory activity of Chithiramoola Rasayanam.

The drug Chithiramoola Rasayanam has been selected for this study to evaluate its efficacy on the Anti-inflammatory activity and Analgesic activity in the management of Soolai.

Biochemical analysis of the drug Chithiramoola Rasayanam reveals the presence of sodium, phosphate , iron, tannic acid and alkaloids.

In the toxicological studies, the drug does not exhibit any mortality up to the dose of 5gm/kg (p.o.)

In the pharmacological studies the drug Chithiramoola Rasayanam exhibits significant Anti-inflammatory and Analgesic activity against low and higher doses.

From the clinical study 95% of patients relieved from pain, 89% of patients relieved from swelling, 71% of patients relieved from stiffness , 50% of patients relieved from crepitus , 85% of patients relieved from restricted movements and no adverse effects were observed during trial period**.**

From the statistical analysis-paired 't' test, the drug Chithiramoola Rasayanam is statistically significant.

Statistically, the paired 't' test shows statistical significance for symptoms before and after the treatment.($p<0.0001$)

The drug Chithiramoola Rasayanam has

- Anti-inflammatory and Analgesic Activity.
- No reported side effects
- No undoing effects
- Encouraging clinical results.

From the clinical and statistical analysis it is proved that the drug Chithiramoola Rasayanam is statistically significant on activity in the management of Soolai .

CONCLUSION

- The literature and research journal review of the herbo drug shows that it has Anti-inflammatory activity.
- The safety studies (acute toxicity and sub-oral toxicity) studies conducted revealed that the trial drug Chithiramoola Rasayanam is safe. There were no abnormalities found in blood investigation and histo-pathological examination. Hence it can be reasonably assumed that the drug is safe for human use.
- The pharmacological study conducted in animal model shows significant Anti-inflammatory activity.

Clinical study revealed the therapeutic efficacy of the trial drug by relieved from pain, patients relieved from swelling, relieved from stiffness, 85% of patients relieved from restricted movements and no adverse effects were observed during trial period**.**

- There were no adverse reactions complained during the clinical trial.

Hence, the drug Chithiramoola Rasayanam can be used in the management of Soolai.

TABLES FOR TRAIL DRUG-1 PALAGARAI PARPAM

QUALITATIVE ANALYSIS:

.S.NO	PARAMETERS	RESULTS
1.	Phosphate	Absent
2.	Sulphate	Absent
3.	Magnesium	Present
4.	Iron	Present
5	Aminoacids	Present
6.	Starch	Absent
7.	Flavonoids	Absent
8.	Proteins	Absent
9.	Tannic acid	Absent
10.	Glycosides	Absent

Trail Drug 1Table -1

PHYSICAL PROPERTIES

S.NO	Characteristic test	Results
1.	pH	9.79
2.	Ash Value	0.03
3.	Water soluble ash	0.06
4.	Acid insoluble ash	0.43

Trail Drug 1 Table -2

Preliminary Acid, Basic Radicals Screening Of Palagarai Parpam (PP)

S.No.	Constituents	PP
1.	Calcium	+
2.	Iron (Ferric)	+
3.	Iron (Ferrous)	+
4.	Chloride	+
5.	Phosphate	_
6.	Potassium	_
7.	Sodium	+
8.	Sulphate	_

Trail Drug 1Table -3

METAL CONTENT:

SAMPLE NAME	Fe (ppm)	Zn (ppm)	K (ppm)	Na (ppm)	Ca (ppm)	Mg (ppm)
PALAGARAI PARPAM	ND	0.055	_	_	0.021	0.35

Trail Drug 1Table -4

TOXICOLOGY TABLES-

Trial Drug1 Table 5: Dose finding experiment and its behavioural Signs of Toxicity

No	Dose mg/kg	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
1.	**250**	+	-	-	+	-	+	-	+	-	-	-	-	-	-	-	+	-	-	-	-
2	**500**	+	+	-	+	-	+	-	+	-	-	-	-	-	-	+	+	+	-	-	-
3	**1000**	+	+	-	+	-	+	+	+	+	+	-	-	-	-	+	+	+	+	+	-
4	**2000**	+	+	-	+	-	+	+	+	+	+	-	-	+	+	+	+	+	+	+	+

1. Alertness 2. Aggressiveness 3. Pile erection 4. Grooming 5. Gripping 6. Touch Response 7. Decreased Motor Activity 8. Tremors 9. Convulsions 10. Muscle Spasm 11. Catatonia 12. Muscle relaxant 13. Hypnosis 14. Analgesia 15.Lacrimation 16. Exophthalmos 17. Diarrhoea 18. Writhing 19. Respiration 20. Mortality

Trial Drug1 Table 6. Body wt (g) of rats exposed to Palagarai Parpam for 28days.

Dose(mg/kg/day)	Days				
	1	7	14	21	28
Control	166.82±1.48	167.81±1.03	167.91±1.77	168.16±1.92	168.97±1.06
25	193.28±0.93**	193.74±0.71**	194.06±0.91**	195.00±0.67**	195.74±0.74**
50	203.53±3.11**	204.02±3.44**	204.50±3.46**	205.32±2.87**	205.73±2.61**
100	208.88±1.13**	207.57±1.47**	204.80±1.33**	208.74±0.57**	208.43±0.51**

Values are mean ± S.E.M. (Dunnet 't' test). *P<0.05; **P<0.01. N=6.

Trial Drug1 Table 7. Food (g/day) intake of rats exposed to *Palagarai Parpam* for 28days.

Dose (mg/kg/day)	Days (gms/rats)				
	1	7	14	21	28
Control	44.05±2.99	44.88±2.15	46.14±2.12	45.08±2.55	47.52±3.21
25	42.27±2.78	45.33±2.46	46.46±2.66	49.10±2.96	48.15±3.00
50	40.38±2.19	42.04±2.59	44.62±2.47	45.86±3.16	47.00±3.08
100	43.61±2.56	45.28±2.83	46.11±2.85	50.17±2.00	50.00±3.13

Values are mean ± S.E.M. (Dunnet 't' test). *P<0.05; **P<0.01. N=6.

Trial Drug 1Table 8. Water (ml/day) intake of rats exposed to Palagarai Parpam for 28days.

Dose (mg/kg/day)	Days(ml/rat)				
	1	**7**	**14**	**21**	**28**
Control	55.00±2.89	52.07±3.38	55.28±3.18	52.37±3.10	51.20±3.29
25	52.18±2.46	50.22±3.00	45.23±4.00	46.15±3.04	40.55±2.88
50	49.74±2.88	40.16±3.78	40.87±3.39*	42.10±2.98	44.10±3.27
100	52.31±3.50	54.67±3.05	51.26±3.88	48.20±3.10	52.23±3.65

Values are mean ± S.E.M. (Dunnet 't' test). *P<0.05; **P<0.01. N=6.

Tria Drug l Table 9. Hematological parameters after 28days treatment with *Palagarai Parpam* in rats.

Parameter	Control	25 mg/kg	50 mg/kg	100 mg/kg
RBC ($X10^6$/µL)	8.20±0.63	8.72±0.80	9.22±1.10	9.03±0.52
HB (%)	14.16±1.24	15.10±1.22	15.84±0.99	15.12±1.18
Leukocyte ($X10^3$/µL)	7.37±1.32	7.50±1.31	6.89±0.93	6.62±0.54
Platelets ($X10^3$/µL)	840.4±80.42	816.25±87.74	837.1±87.14	780.76±70.32
MCV (gl)	51.24±5.08	50.16±4.32	51.60±5.41	56.14±4.86
N	23.22±2.16	22.53±3.70	24.74±3.62	25.18±3.11
L	69.82±6.15	67.46±6.55	66.24±6.28	66.12±3.18
M	2.0±0.34	2.0±0.35	2.24±0.28	2.32±0.26
E	1.42±0.54	1.61±0.60	1.28±0.42	1.00±0.11
B	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
ESR(mm)	1±00	1±00	1±00	1±00
PCV	42.10±1.02	44.6±3.7	44.04±2.2	45±1.8

Values are mean ± S.E.M. (Dunnett's test). *P<0.05; **P<0.01. N=6.

Trial Drug1 Table 10. Effect of *Palagarai Parpam* biochemical parameters.

Dose (mg/kg)	Control	25 mg/kg	50 mg/kg	100 mg/kg
Total Bilirubin (µmol/l)	17.12±2.16	16.70±1.10	15.12±1.15	20.66±1.35
Bilirubin direct (µmol/l)	13.64±1.21	11.74±1.04	10.78±0.86	10.12±0.73*
ALP (U/L)	60.11±3.12	85.11±2.62**	102.10±1.92**	105.45±2.31**
SGOT (U/L)	84.02±6.2	90.16±8.45	87.52±6.31	96.2±7.52
SGPT(U/L)	71.72±7.2	74.31±6.2	82.2±7.10	93.5±7.50
Total Protein(g/dl)	8.21±0.25	7.55±0.23	7.55±0.72	7.18±0.61
Albumin(g/dl)	2.62±0.18	2.54±0.17	2.52±0.16	2.65±0.10
Globulin(g/dl)	6.82±0.28	5.18±0.28**	4.78±0.22**	4.73±0.30**
Urea(mg/dL)	6.35±0.25	7.30±0.19*	7.58±0.25**	8.85±0.22**
Creatinine (mg/dL)	0.17±0.04	0.22±0.05	0.50±0.05**	0.52±0.04**
Uric acid (mg/dL)	1.92±0.07	2.12±0.08	1.39±0.08**	1.29±0.07**
Na m.mol	148.00±1.10	147.36±1.12	146.28±0.54	145.20±0.50
K m.mol	6.12±0.88	5.90±1.00	5.68±0.78	5.82±0.69
Cl m.mol	100.05±4.26	101.27±5.51	99.82±4.72	100.14±5.10

Values are mean ± S.E.M. (Dunnett's test). *P<0.05; **P<0.01. *Vs. control*

Trial Drug 1Table-11. Lipid Profile

Dose (mg/kg)	Control	25 mg/kg	50 mg/kg	100 mg/kg
Total cholestrol(mg/dL)	75.28±4.4	78.10±7.6	82.20±8.10	82.64±6.2
HDL(mg/dL)	114.5±0.47	161.57±0.38**	150.39±0.49**	140.02±0.37**
LDL(mg/dL)	32.40±4.71	95.62±0.64	71.70±3.52**	86.43±0.57**
VLDL(mg/dl)	16.32±2.42	15.27±2.24	16.10±1.44	15.06±1.11
Triglycerides (mg/dl)	66.56±1.50	83.29±1.04**	118.30±1.02**	113.15±1.05**
Blood glucose(mg/dl)	90.77±0.48	81.16±0.32**	58.56±0.45**	52.00±1.00**

Values are mean ± S.E.M. (Dunnett's test). *P<0.05; **P<0.01. *Vs. control*

Trial Drug1*table-12 Urine Analysis*

Parameters	**Control**	**25 mg/kg**	**50 mg/kg**	**100 mg/kg**
Colour	Yellow	Yellow	Yellow	Yellow
Transparency	Clear	Slightly turbid	Slightly cloudy	Slightly turbid
Specific gravity	1.010	1.010	1.010	1.010
PH	>7.2	>8.0	>8.0	>9.0
Protein	Nil	3+	3+	3+
Glucose	Nil	Nil	Nil	Nil
Bilirubin	-ve	-ve	-ve	-ve
Ketones	-ve	+ve	+ve	+ve
Blood	Absent	Absent	Absent	Absent
Urobilinogen	Normal	Abnormal	Abnormal	Abnormal
Pus cells	0-cells/HPF	1-cell/HPF	2-cells/HPF	1-cell/HPF
RBCs	Nil	Nil	0-1cells/HPF	Nil
Epithelial cells	Nil	1-cell/HPF	Nil	1-cell/HPF
Crystals	Nil	Nil	Nil	Nil
Casts	Nil	Nil	Nil	Nil
Others	Bacteria seen	Bacteria seen	Bacteria seen	Bacteria seen

Trial Drug 1Table 12. Effect of Palagarai Parpam on organ weight

Dose (mg/kg)	Control	25 mg/kg	50 mg/kg	100 mg/kg
Brain	1.96±0.03	1.81±0.04**	1.90±0.03	1.91±0.02
Lungs	1.74±0.05	1.75±0.12	1.69±0.05	1.61±0.05
Heart	1.32±0.05	1.17±0.04	1.25±0.05	1.32±0.04
Liver	8.72±0.30	8.41±0.27	8.73±0.34	8.76±0.40
Pancreas	1.38±0.09	1.40±0.12	1.60±0.10	1.66±0.12
Spleen	0.91±0.03	0.79±0.03	0.86±0.04	0.92±0.06
Adrenals	0.03±0.00	0.04±0.00	0.03±0.02	0.03±0.00
Kidneys	1.27±0.03	1.16±0.02*	1.25±0.03	1.32±0.03
Ovary	0.08±0.01	0.09±0.00	0.06±0.03	0.07±0.00
Uterus	0.87±0.05	0.75±0.06	0.72±0.06	0.60±0.04**

Values are mean ± S.E.M. (Dunnet 't' test). *P<0.05; **P<0.01 *Vs control* N=6.

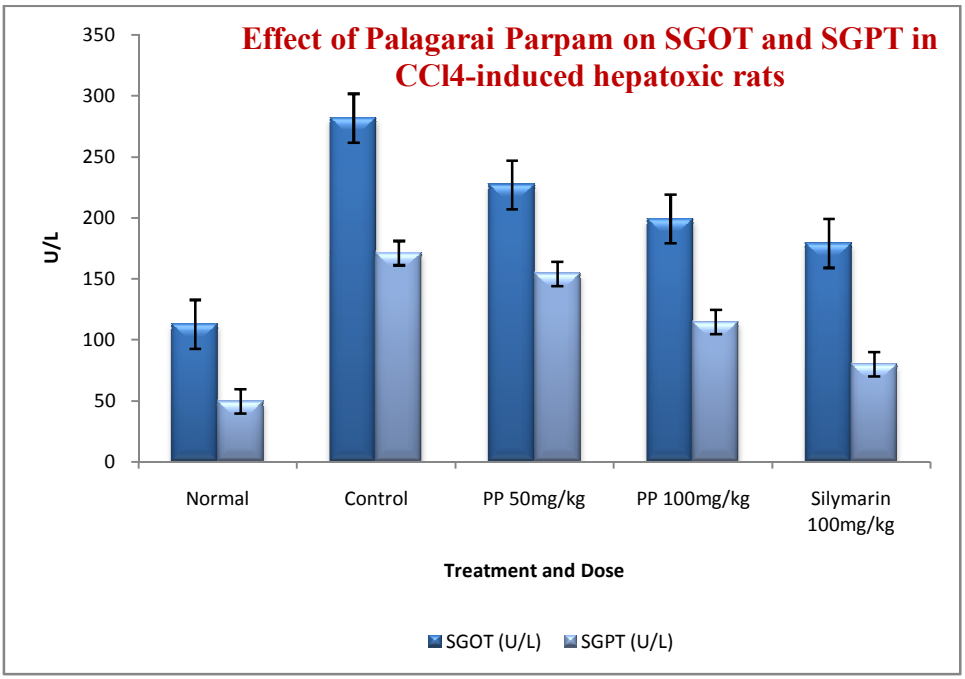
PHARMACOLOGICAL TABLES

Trial Drug 1Table 13. Effect of Palagarai Parpam on CCl4-induced hepatoxic rats.

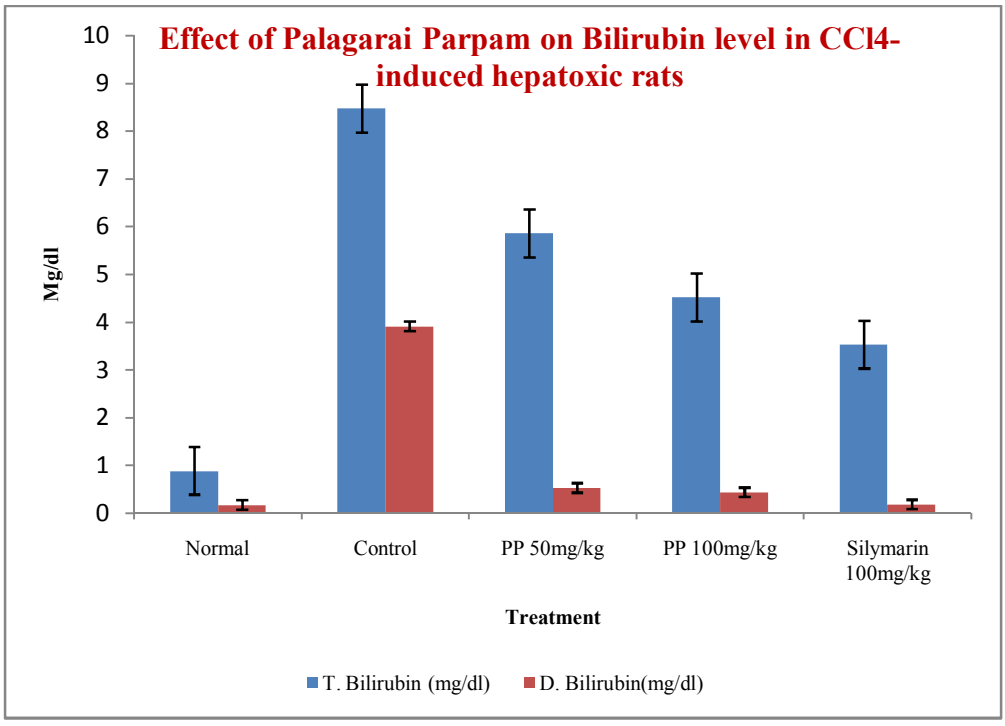
Group	SGOT (U/L)	SGPT (U/L)	ALP (mg/dl)	T. Bilirubin (mg/dl)	D.Bilirubin(mg/dl)
Group I	112.9±0.71	49.5±0.05	217.6±0.12	0.890±0.001	0.179±0.001
Group II	281.9±0.61**	171.2±0.60**	890.8±0.57**	8.47±0.061**	3.91±0.006**
Group III	227.2±1.18**	154.2±0.63**	520±0.64**	5.86±0.577**	0.532±0.002**
Group IV	199.2±0.51**	114.6±1.80**	669.7±0.55**	4.52±0.105**	0.439±0.001**
Group V	179.1±0.46**	80.1±0.55**	398.1±1.07**	3.53±0.115**	0.185±0.002ns

Values are the Mean ± SEM of six rats/ treatment.

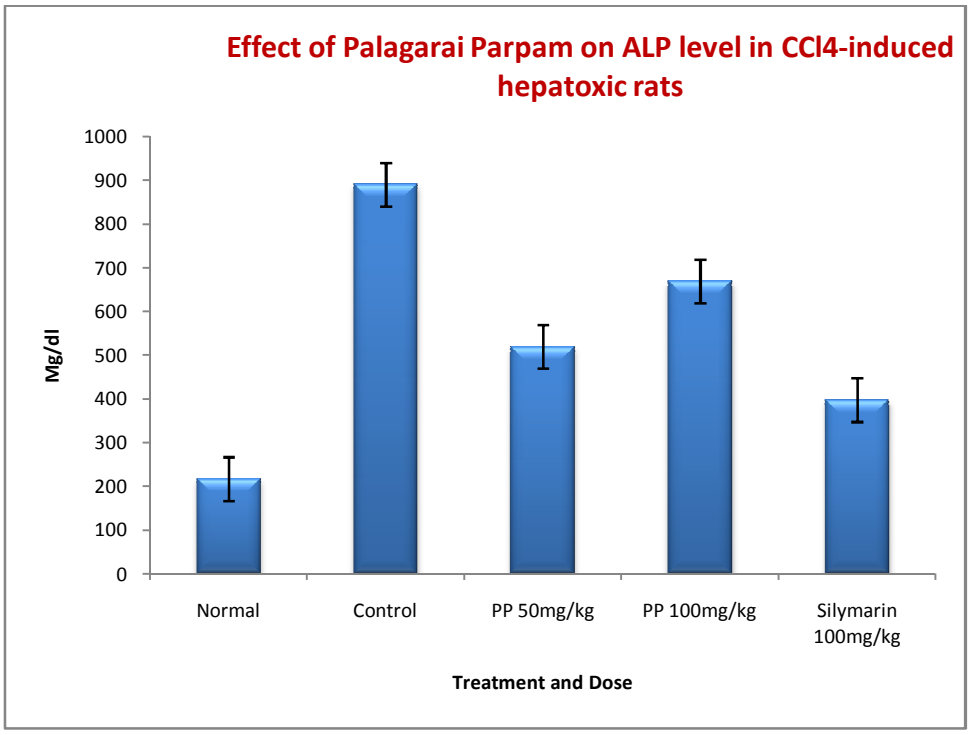
Trial Drug 1 Chart 1



Trial Drug 1 Chart 2



Trial Drug 1 Chart 3



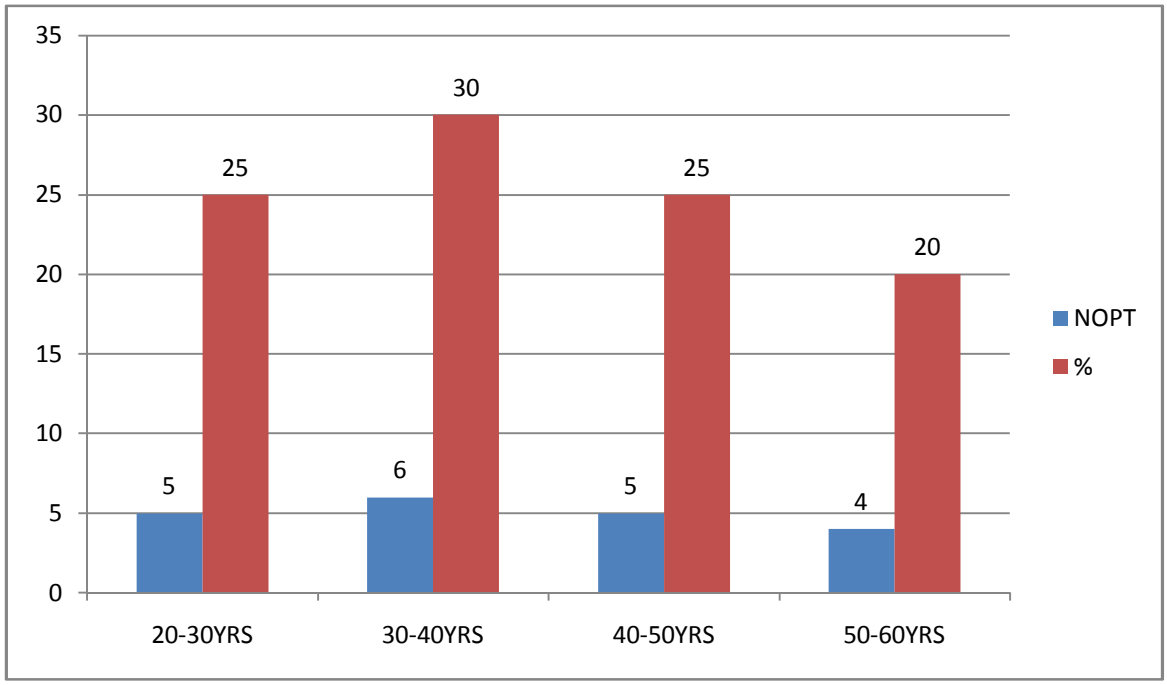
TRAIL DRUG 1

CLINICAL ASSESMENT

AGE DISTRIBUTION

AGE	NO OF PATIENTS	PERCENTAGE
20-30Yrs	5	25
30-40 yrs	6	30
40-50 yrs	5	25
50-60yrs	4	20

AGE DISTRIBUTION

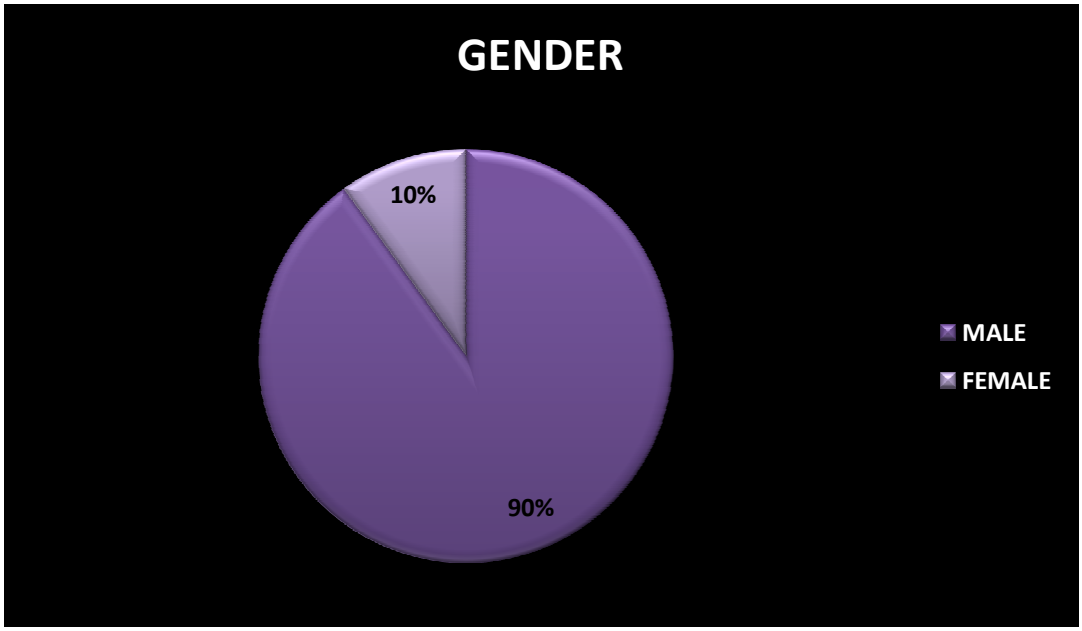


Nopt- Number of Patients

GENDER DISTRIBUTION

S.NO	GENDER	NO.OF PATIENTS	PERCENTAGE
1.	FEMALE	2	10%
2.	MALE	18	90%

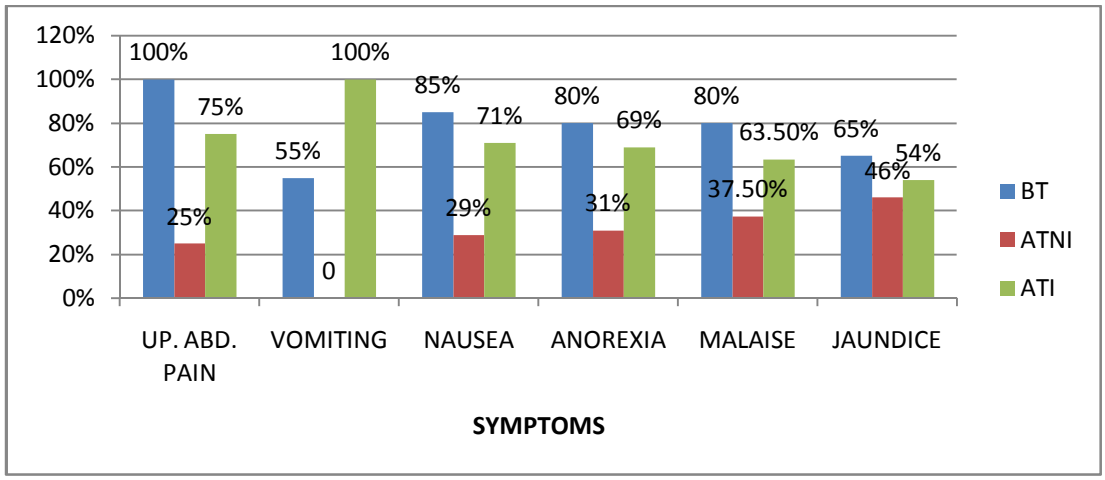
GENDER DISTRIBUTION



IMPROVEMENT SHOWING SIGNS AND SYMPTOMS BEFORE AND AFTER TREATMENT OF KALLERAI NOI.

SYMPTOMS	NO. OF PATIENTS WITH SYMPTOMS			
	BT	AFTER TREATMENT		IMPROVEMENT PERCENTAGE
		NO IMPROVEMENT	IMPROVEMENT	
UPPER ABDOMEN PAIN	20 (100%)	5(25%)	15 (75%)	75%
VOMITING	11(55%)	0	11 (100%)	100%
NAUSEA	17(85%)	5(29%)	12 (71%)	71%
ANOREXIA	16(80%)	5(31%)	11 (69%)	69%
MALAISE	16(80%)	6(37.5%)	10(62.5%)	63.5%
JAUNDICE	13(65%)	6(46%)	7(54%)	54%

IMPROVEMENT SHOWING SIGNS AND SYMPTOMS BEFORE AND AFTER TREATMENT OF KALLERAI NOI.



BT- BEFORE TREATME

AT- AFTER TREATMENT
AT - AFTER TREATMENT NO IMPROVEMENT

STATISTICAL ANALYSIS:

All collected data were entered into MS Excel software using different columns as variables and rows as patients. SPSS software was used to perform statistical analysis. Basic descriptive statistics include frequency distributions and cross-tabulations were performed. The quantity variables were expressed as Mean ± Standard Deviation and qualitative data as percentage. A probability value of <0.05 was considered to indicate as statistical significance. Paired 't' test was performed for determining the significance between before and after treatmen

DRUG 1

KALLERAL NOI

Trail Drug 1 Table 13

Paired t test for Symptoms before and after treatment:

Variable	Obs	Mean	Std.dev	t.value	P value
Before symptoms	20	4.80	0.894	-10.031	P<0.0001
After symptoms	20	1.35	1.894		

For symptom of mean± standard deviation before treatment is 4.80 ± 0.894 and after treatment is 1.35 ± 1.894 which is statistically significant(p<0.0005).

Trail Drug 1 Table 14

Paired t test for total bilirubil before and after treatment:

Total bilirubin	Obs	Mean	Std.dev	t.value	P value
Before symptoms	20	.2041	1.884	2.871	P<0.01
After symptoms	20	1.245	.8678		

For Total bilirubin of mean± standard deviation before treatment is .2041and after treatment is 1.245which is statistically significan t (p<0.01) .Paired t test for before and after treatment:

Trail Drug 1 Table 15

Paired t test for SGOT,SGPT before and after treatment:

Variable	Obs	Mean		Std.dev		t.value		P value	
		SGOT	SGPT	SGOT	SGPT	SGOT	SGPT	SGOT	SGPT
Before symptoms	20	48.80	43.80	43.85	29.17	2.433	2.170	P<0.025	P<0.049
After symptoms	20	26.12	31.40	15.08	14.255				

For SGOT of mean± standard deviation before treatment is 48.80± 43.80 and after treatment is 26.12 ± 31.40 which is statistically significant($p<0.025$).

For SGPT of mean± standard deviation before treatment is 43.80 ± 29.17 and after treatment is 31.40 ± 14.255 which is statistically significant($p<0.049$).

IMPROVEMENT IN PROGNOSIS OF SYMPTOMS OF KALLERAL NOI

S.NO	OP/IP	AGE	SEX	MALAISE		ANOREXIA		NAUSEA		VOMITING		JAUNDICE		UPPER ABDOMEN PAIN	
				BT	AT		AT	BT	AT	BT	AT	BT	AT	BT	AT
1	C80708	29	M	+	_	+	_	+	_	-	_	_	_	+	_
2	C84096	44	F	+	+	+	+	_	_	_	_	_	_	+	_
3	C84788	32	M	+	+	+	+	+	_	+	_	+	+	+	+
4	C87981	44	M	+	_	+	_	+	+	_	_	+	+	+	_
5	C75157	46	M	+	_	+	_	+	_	_	_	+	_	+	_
6	C88089	25	M	+	+	+	_	+	_	+	_	+	_	+	_
7	5005	55	M	+	+	+	+	+	+	_	_	+	+	+	+
8	C87693	26	M	_	_	_	_	+	_	_	_	+	_	+	_
9	c88638	35	M	+	_	+	_	+	+	+	_	+	+	+	+
10	c91112	60	M	_	_	+	_	+	+	_	_	+	+	+	+
11	C91323	60	F	+	_	_	_	+	_	+	_	_	_	+	_
12	C90680	29	M	+	+	+	_	+	_	+	_	_	_	+	_
13	C91558	60	M	+	_	+	_	+	_	_	_	_	-	+	_
14	C92972	43	M	_	-	+	-	+	-	+	-	+	_	+	_
15	C92771	26	M	+	+	+	_	+	_	+	_	_	_	+	_
16	5087	34	M	-	_	+	+	+	_	+	_	+	+	+	+
17	C94608	30	M	+	_	+	+	+	_	_	_	+	_	+	_
18	C13733	30	M	+	_	+	_	_	_	+	_	_	_	+	_
19	DOO9130	49	M	+	_	_	_	+	_	+	_	+	_	+	_
20	D008583	39	M	+	_	+	_	+	+	+	_	+	_	+	_

SPECIFIC INVESTIGATION FOR KALLRRAL NOI

S.NO	OP/IP	AGE	SEX	BTFBS	ATFBS	BTPBS	ATPBS	BTTB	ATTB	BTDB	ATDB	BTIB	ATIB	BTOT	ATOT	BTPT	ATPT	BTALP	ATALP	BTTP	ATTP	BTALB	ATALB	BTGLO	ATGLO	BTUBS	ATUBS	BTUBP	ATUBP
1	C80708	29	M	100	90	140	110	0.6	0.6	0.2	0.2	0.4	0.4	55	42	49	58	260	270	4.9	6.8	3.5	4.1	2.1	2.8	_	_	_	_
2	C84096	44	F	101	89	141	111	0.4	0.3	0.2	0.3	0.2	0.1	40	13	41	15	177	150	6.7	7.1	3.3	2.1	2.4	5	_	_	_	_
3	C84788	32	M	100	89	140	111	2.9	1.3	2.3	0.8	0.6	0.1	22	34	23	33	189	232	7.1	6.5	5.1	3.6	2	2.9	+	_	+	_
4	C87981	44	M	96	90	116	106	1.6	0.4	1.2	0.2	0.4	0.4	21	21	22	24	166	214	6.9	6.2	4.9	3.6	2	2.6	_	_	_	_
5	C75157	46	M	110	100	140	129	2	1.5	0.7	0.5	1.3	1	22	20	25	21	186	194	7	7	5	4	2	3	_	_	_	_
6	C88089	25	M	82	80	100	90	1.5	0.7	0.7	0.5	0.8	0.2	10	5	11	10	145	140	7.3	7.1	5.2	5	2.1	2.1	_	_	_	_
7	5005	55	M	85	90	147	110	5.1	3	2	1	3.1	2	17	10	18	15	168	150	7.4	7.4	4.6	4.6	2.8	2.8	+	+	+	+
8	C87693	26	M	74	104	90	121	3.5	1.9	1.1	0.6	2.4	1.3	78	16	100	17	240	140	7.8	5.9	4.9	3	2.9	2.9	_	_	-	_
9	c88638	35	M	101	117	117	140	1.5	2	1.2	1.8	0.3	0.2	28	35	32	40	185	200	6.9	6.5	4.8	4.5	2.1	2	+	_	+	+
10	c91112	60	M	90	115	134	200	1.7	2	1.3	1.1	0.4	0.9	24	30	26	40	220	133	6.9	7.2	4	4.8	2.9	2.4	+	+	+	+
11	C91323	60	F	110	94	130	104	0.5	0.7	0.2	0.3	3	0.4	45	40	46	36	149	155	6.9	7	4.2	4	2.3	3	_	_	_	_
12	C90680	29	M	94	90	120	110	0.5	0.7	0.2	0.5	0.3	0.2	127	46	124	60	290	180	7.4	7	5.1	5	2.3	2	_	_	_	_
13	C91558	60	M	120	104	159	149	0.5	0.7	0.2	0.3	0.3	0.4	40	22	43	24	199	171	5.5	6.5	3.2	3.6	2.3	2.9	_	_	_	_
14	C92972	43	M	98	96	114	118	1.9	1.2	0.8	0.5	1.1	0.7	22	27	27	32	87	130	6.7	6.4	3.8	4	2.9	2	_	_	_	_
15	C92771	26	M	80	90	110	120	0.3	0.4	0.1	0.2	0.2	0.2	32	20	33	21	184	200	5.1	7.5	3.1	4.7	2	2.8	_	_	_	_
16	5087	34	M	90	83	120	110	2.9	1.9	0.9	0.9	2	1	40	30	50	43	180	200	6.6	7	4.5	4.5	2.1	2.5	_	_	_	_
17	C94608	30	M	82	103	125	145	1.9	1.2	0.5	0.5	1.4	0.7	14	20	16	21	155	216	6.4	6.5	4	4.2	2.4	2.3	_	_	_	_
18	C13733	30	M	90	103	140	121	0.5	0.4	0.2	0.2	0.3	0.2	36	26	64	38	260	297	6.6	5.7	4.1	3	2.5	2.7	_	_	_	_
19	DOO9130	49	M	104	100	118	110	2.9	0.7	1.2	0.3	1.7	0.4	156	40	82	30	207	41	5.9	5.2	3.7	3.2	2.2	2	_	_	+	_
20	D008583	39	M	93	90	153	140	8	3.3	4.4	1.9	3.6	1.4	147	65	44	50	231	200	5.8	5.1	3.2	3.2	2.6	2.5	+	+	+	+

AFTER TREATMENT INVESTIGATION OF KALLERAL NOI

S.NO	OP/IP NO	AGE	SEX	ATHB	ATTC	ATDCP	ATDCL	ATDCE	ATDCM	ATHESR	ATOESR	ATPLT	ATUREA	ATCRE	ATCHO	ATHDL	ATLDL	ATVLDL	AUALB	AUFS	AUPS	ATBS	ATBP	urobilinogen	PUSCELLS	EPI.CELLS
1	C80708	29	M	15.8	9100	70	23	7	0	2	10	1.8	15	0.4	128	24	88	37	NIL	NIL	NIL	NIL	NEG	NML	1-2P	1-2E
2	C84096	44	F	12.6	8000	65	30	5	0	10	20	2.4	14	4	146	30	92	98	NIL	NIL	NIL	NIL	NEG	NML	1-2P	1-2E
3	C84788	32	M	17.3	8300	50	47	3	0	2	4	14	0.4	157	32	79	56	280	NIL	NIL	NIL	NIL	NEG	NML	1-2P	1-2E
4	C87981	44	M	16.6	7500	60	37	3	0	2	4	2.2	14	0.4	121	28	100	31	NIL	NIL	NIL	NIL	NEG	NML	1-2P	1-2E
5	C75157	46	M	15.4	5600	49	46	5	0	2	8	2.1	22	0.8	192	32	100	25	NIL	NIL	NIL	NIL	NEG	NML	1-2P	1-2E
6	C88089	25	M	16.5	10000	70	30	0	0	2	4	2.5	14	0.5	200	40	100	25	NIL	NIL	NIL	NIL	NEG	NML	1-2P	1-2E
7	5005	55	M	11	6000	70	25	5	0	22	40	1.2	16	0.3	97	40	90	14	NIL	NIL	NIL	present	POSITIVE	NML	2-4P	2-4E
8	5016	26	M	14.9	7400	65	30	5	0	2	4	2.6	14	0.4	144	38	94	35	NIL	NIL	NIL	NIL	NEG	NML	1-2P	1-2E
9	c88638	35	M	15	6900	70	28	2	0	2	8	2.2	20	0.6	200	38	100	40	NIL	NIL	NIL	NIL	NEG	NML	1-2P	1-2E
10	c91112	60	M	14.2	10300	70	24	6	0	4	16	3.1	30	0.9	247	49	131	44	NIL	NIL	NIL	NIL	NEG	NML	1-2P	1-2E
11	C91323	60	F	13.7	7700	65	30	5	0	4	8	2.4	14	0.4	205	42	98	39	NIL	NIL	NIL	NIL	NEG	NML	1-2P	1-2E
12	C90680	29	M	16.5	1000	70	30	0	0	2	4	2.5	14	0.5	200	40	100	25	NIL	NIL	NIL	NIL	NEG	NML	1-2P	1-2E
13	C91558	60	M	15.6	5300	66	29	5	0	2	4	2.5	22	0.7	159	42	80	66	NIL	NIL	NIL	NIL	NEG	NML	1-2P	1-2E
14	C92972	43	M	14.9	7000	55	39	5	0	2	6	2.6	17	0.6	196	36	26	133	NIL	NIL	NIL	NIL	NEG	NML	1-2P	1-2E
15	C92771	26	M	15.2	5600	61	30	9	0	2	6	2.9	14	0.4	160	32	80	15	NIL	NIL	NIL	NIL	NEG	NML	1-2P	1-2E
16	5087	34	M	17.8	5600	60	30	10	0	4	6	2.2	16	2.2	280	50	140	50	NIL	NIL	NIL	NIL	NEG	NML	1-2P	1-2E
17	C94608	30	M	12.2	6200	55	41	4	0	2	4	2.3	16	0.5	177	36	92	30	NIL	NIL	NIL	NIL	NEG	NML	1-2P	1-2E
18	C13733	30	M	12.9	8700	60	36	4	0	2	4	2.5	18	0.6	214	40	101	21	NIL	NIL	NIL	NIL	NEG	NML	1-2P	1-2E
19	DOO9130	49	M	13.3	7000	71	22	7	0	2	6	1.9	22	0.8	175	35	88	19	NIL	NIL	NIL	NIL	NEG	NML	1-2P	1-2E
20	D008583	39	M	11.6	9500	70	25	5	0	10	20	2	16	0.6	145	40	100	85	NIL	NIL	NIL	NIL	NEG	NML	1-2P	1-2E

BEFORE TREAT MENT INVESTIGATIONS OF KALLERAL NOI.

S.NO	OP/IP	AGE	SEX	HB mg/dl	TC Cells/cu. mm	DC %				ESR mm		PLT Cells/cu.mm	UREA mg/dl	CRE mg/dl	CHO mg/dl	HDL mg/dl	LDL mg/dl	VLDL mg/dl	HBsAg	UALB	UFS	UPS	BS	BP	PUSCELLS	Epi
						P	L	E	M	1\2HR	1hr															
1	C80708	29	M	15	11300	67	28	5	0	16	32	2	14	0.4	147	32	94	18	NEG	NIL	NIL	NIL	NIL	NEG	1-2P	1-2 E
2	C84096	44	F	13	9100	70	27	30	0	8	18	2.9	22	0.6	194	26	145	287	NEG	NIL	NIL	NIL	NIL	NEG	1-2P	1-2 E
3	C84788	32	M	16	8600	40	52	8	0	2	10	3.3	20	0.7	177	34	106	42	NEG	NIL	NIL	NIL	NIL	NEG	1-2P	1-2E
4	C87981	44	M	17	7800	64	31	5	0	8	16	2	19	0.6	146	28	50	18	NEG	NIL	NIL	NIL	NIL	NEG	1-2P	1-2 E
5	C75157	46	M	16	5900	54	41	5	0	2	6	2.2	28	14	134	33	64	17	NEG	NIL	NIL	NIL	NIL	NEG	1-2P	1-2E
6	C88089	25	M	17	9600	70	25	5	0	2	6	2.9	14	0.4	275	45	218	59	NEG	NIL	NIL	NIL	NIL	NEG	1-2P	1-2E
7	5005	55	M	9.8	4000	62	30	8	0	44	88	0.8	16	0.5	97	20	108	22	NEG	NIL	NIL	NIL	PoS	pre	2-4P	2-4E
8	5016	26	M	14	6100	50	38	12	0	2	6	2.2	33	1	193	30	130	34	NEG	NIL	NIL	NIL	NIL	NEG	1-2P	1-2E
9	c88638	35	M	14	6600	54	42	4	0	8	18	3.2	14	0.1	227	35	195	44	NEG	NIL	NIL	NIL	NIL	NEG	1-2P	1-2E
10	c91112	60	M	14	8000	62	34	4	0	22	4	32	24	0.9	184	39	96	50	NEG	NIL	NIL	NIL	NIL	NEG	1-2P	1-2E
11	C91323	60	F	13	7300	65	30	5	0	6	16	2.5	16	0.5	217	35	139	46	NEG	NIL	NIL	NIL	NIL	NEG	1-2P	1-2E
12	C90680	29	M	17	9600	70	25	5	0	2	6	2.9	14	0.4	275	45	218	59	NEG	NIL	NIL	NIL	NIL	NEG	1-2P	1-2E
13	C91558	60	M	15	5900	64	29	6	1	6	14	2.6	18	0.5	172	42	123	98	NEG	NIL	NIL	NIL	NIL	NEG	1-2P	1-2E
14	C92972	43	M	18	8600	64	30	6	0	12	28	2.6	15	0.5	179	35	116	35	NEG	NIL	NIL	NIL	NIL	NEG	1-2P	1-2E
15	C92771	26	M	15	4700	62	36	2	0	2	4	2.7	20	0.6	159	32	119	17	NEG	NIL	NIL	NIL	NIL	NEG	1-2P	2-3E
16	5087	34	M	23	12600	86	12	2	0	2	4	1.1	20	0.7	360	30	17	17	NEG	NIL	NIL	NIL	NIL	NEG	1-2P	1-2E
17	C94608	30	M	15	7300	60	35	5	0	2	4	2.2	15	0.4	182	40	104	32	NEG	NIL	NIL	NIL	NIL	NEG	1-2P	1-2E
18	C13733	30	M	13	8500	60	35	5	0	6	12	5.1	18	0.6	183	36	64	49	NEG	NIL	NIL	NIL	NIL	NEG	1-2P	1-2E
19	DOO9130	49	M	16	11200	84	12	4	0	4	1	1.1	14	0.5	308	49	139	22	NEG	NIL	NIL	NIL	NIL	NEG	1-2P	1-2E
20	D008583	39	M	11	11000	70	24	6	0	14	30	2.4	14	0.4	248	45	112	28	NEG	NIL	NIL	NIL	Pos	PRE	2-4P	1-2E

PROGNOSIS BEFORE AND AFTER TREATMENT

S.NO	OP/IP	USG SCAN	
		BT	AT
1	C80708	Fatty liver	Normal study
2	C84096	Fatty liver	Normal study
3	C84788	Fatty liver	Normal study
4	C87981	Fatty liver	Fatty liver
5	C75157	Fatty liver	Normal Study
6	C88089	Normal study	Normal study
7	5005	Fatty liver	Fatty liver
8	C87693	Normal study	Normal study
9	c88638	Normal study	Normal study
10	c91112	Fatty liver	Fatty liver
11	C91323	Fatty liver	Fatty liver
12	C90680	Normal study	Normal study
13	C91558	Normal study	Normal study
14	C92972	Normal study	Normal study
15	C92771	Normal study	Normal study
16	5087	Normal study	Normal study
17	C94608	Fatty liver	N ormal study
18	C13733	Normal study	Normal study
19	DOO9130	Normal study	Normal study
20	D008583	Fatty liver	Normal study

INDIAN SCAN
ADVANCED DIAGNOSTIC CENTRE
◆ Multi Channel MRI ◆Multi Slice CT ◆ Digital Color Doppler ◆ Digital Ultrasonography
◆ Echocardiography ◆Computerised ECG ◆ Treadmill ◆PFT ◆ Endoscopy ◆Digital X-Ray ◆Laboratory

Patient Name:	*Mr. MURTHY*	*Age/Sex:*	*47/M*
Referred by:	*Dr.Prabhapathy*	*Visit Date:*	*19/07/2012*

ABDOMEN & KUB SCAN REPORT

Liver : *Liver shows diffuse abnormal increased echoreflectivity . Focal fat sparing noted in the right lobe no abscess or mass lesion seen. CBD AND IHBR appear normal Liver measurement -14.6cms.*

Gall bladder : *Gall Bladder appeared normal. No calculus seen in gall bladder. CBD Appeared normal. No calculus seen in CBD.*

Pancreas : *Pancreas appeared normal.*

Spleen : *Spleen appeared normal. Spleen measured 10.5 cms*

Aorta : *Aorta appeared normal. No Para aortic nodes seen.*

Peritoneal cavity: *Peritoneal cavity appeared normal.*

Adrenals : *Adrenals appeared normal.*

KUB

RK : *Right Kidney measured 10.1 cms. Cortex and collecting system of right kidney*

Appeared normal. No calculi seen.

LK : *Left Kidney measured 9.8 cms. Cortex and collecting system of left kidney Appeared normal. No calculi seen.*

BLADDER : *Bladder appeared normal.*

PROSTATE : *Prostrate appeared normal. Measured in 3.2 X 2.8 X 2.2 cms (Weight = 15.1 GMS).No intra vesicle enlargement of prostate gland seen.*

IMPRESSION

Fatty liver .No other abnormalities seen

DR. NISCHAL., MD., RD
Consultant Radiologist.

INDIAN SCAN
ADVANCED DIAGNOSTIC CENTRE
◆ Multi Channel MRI ◆Multi Slice CT ◆ Digital Color Doppler ◆ Digital Ultrasonography
◆ Echocardiography ◆Computerised ECG ◆ Treadmill ◆PFT ◆ Endoscopy ◆Digital X-Ray ◆Laboratory

Patient Name:	*Mr. MURTHY*	*Age/Sex:*	*47/M*
Referred by:	*Dr.Prabhapathy*	*Visit Date:*	*1/09/2012*

ABDOMEN & KUB SCAN REPORT

Liver : *Liver shows normal. Focal fat sparing noted in the right lobe no abscess or mass lesion seen. CBD AND IHBR appear normal. Liver measurement-12.6cms*

Gall bladder : *Gall Bladder appeared normal. No calculus seen in gall bladder. CBD Appeared normal. No calculus seen in CBD.*

Pancreas : *Pancreas appeared normal.*

Spleen : *Spleen appeared normal. Spleen measured 10.5 cms*

Aorta : *Aorta appeared normal. No Para aortic nodes seen.*

Peritoneal cavity: *Peritoneal cavity appeared normal.*

Adrenals : *Adrenals appeared normal.*

KUB

RK : *Right Kidney measured 10.1 cms. Cortex and collecting system of right kidney Appeared normal. No calculi seen.*

LK : *Left Kidney measured 9.8 cms. Cortex and collecting system of left kidney Appeared normal. No calculi seen.*

BLADDER : *Bladder appeared normal.*

PROSTATE : *Prostrate appeared normal. Measured in 3.2 X 2.8 X 2.2 cms (Weight = 15.1 GMS).No intra vesicle enlargement of prostate gland seen.*

IMPRESSION

Normal study :No other abnormalities seen

DR. NISCHAL., MD., RD
Consultant Radiologist.

MARUTHY HI-TECH
DIAGNOSTIC CENTRE
Govt. Royapettah Hospital Signal, Royapettah, Chennai - 600 014.
Phone : 044 - 28351299, 42105749

Patient Name:	Mr. Nanthagopal	Age/Sex:	29/M
Referred by:	Dr.Prabhapathy	Visit Date:	22/7/2012

ABDOMEN & KUB SCAN REPORT

Liver : **Liver shows normal with duffuse increase in echotexture . No abscess or mass lesion seen. Liver measured – 12.9cms**

Gall Bladder : **Gall Bladder appeared normal. No calculus seen in Gall Bladder. CBD appeared normal. No calculus seen in CBD.**

Pancreas : **Pancreas appeared normal.**

Spleen : **Spleen appeared normal. Spleen measured 9.8cms**

Aorta : **Aorta appeared normal. No Para aortic nodes seen.**

Peritoneal cavity: **Peritoneal cavity appeared normal.**

Adrenals : **Adrenals appeared normal.**

KUB

RK : **Right Kidney measured 9.5 X 3.9 cms. Cortex and collecting System of right kidney appeared normal. No calculi seen.**

LK : **Left Kidney measured 9.7 X 4.1 cms. Cortex and collecting System of left kidney appeared normal. No calculi seen.**

BLADDER : **Bladder appeared normal.**

PROSTATE : **Prostrate appeared normal. Measured in 2.8 X 2.5 X 1.9 cms (Weight = 14.4 gms). No intra vesicle enlargement of prostate gland seen.**

IMPRESSION

FATTTY LIVER .

DR.S.NANDINI AZHAR, MBBS.,DMRD.,
RADIOLOGIST

Dr. S. NANDINI, MBBS., DMRD.,
CONSULTANT RADIOLOGIST
REGN. No. 43367

MARUTHY HI-TECH
DIAGNOSTIC CENTRE
Govt. Royapettah Hospital Signal, Royapettah, Chennai - 600 014.
Phone : 044 - 28351299, 42105749

ABDOMEN & KUB SCAN REPORT

Patient Name:	Mr. Nanthagopal	Age/Sex:	29/M
Referred by:	Dr.Prabhapathy	Visit Date:	4/10/2012

Liver : Liver shows normal. No abscess or mass lesion seen. Liver measured – 12.0cms

Gall Bladder : Gall Bladder appeared normal. No calculus seen in Gall Bladder. CBD appeared normal. No calculus seen in CBD.

Pancreas : Pancreas appeared normal.

Spleen : Spleen appeared normal. Spleen measured 9.8cms

Aorta : Aorta appeared normal. No Para aortic nodes seen.

Peritoneal cavity: Peritoneal cavity appeared normal.

Adrenals : Adrenals appeared normal.

KUB

RK : Right Kidney measured 9.5 X 3.9 cms. Cortex and collecting System of right kidney appeared normal. No calculi seen.

LK : Left Kidney measured 9.7 X 4.1 cms. Cortex and collecting System of left kidney appeared normal. No calculi seen.

BLADDER : Bladder appeared normal.

PROSTATE : Prostrate appeared normal. Measured in 2.8 X 2.5 X 1.9 cms (Weight = 14.4 gms). No intra vesicle enlargement of prostate gland seen.

IMPRESSION

Normal study.

DR.S.NANDINI AZHAR, MBBS.,DMRD.,
RADIOLOGIST

Dr. S. NANDINI, MBBS., DMRD.,
CONSULTANT RADIOLOGIST
REGN. No. 43367

AARTHI SCANS

AARTHI SCANS™

AN ISO 9001 ORGANISATION

ABDOMEN & KUB SCAN REPORT

Patient Name:	Mrs. Saraswathy	Age/Sex:	44/f
Referred by:	Dr.Prabhapathy	Visit Date:	4/7/2012

Liver : Liver is mildly enlarged in size and measures 13.8 cms shows diffuse homogenus increased echo texture.

Gall Bladder: Gall Bladder appeared normal. No calculus seen in Gall Bladder. CBD appeared normal. No calculus seen in CBD.

Pancreas : Pancreas appeared normal.

Spleen : Spleen appeared normal. Spleen measured 9.8cms

Aorta : Aorta appeared normal. No Para aortic nodes seen.

Peritoneal cavity: Peritoneal cavity appeared normal.

Adrenals : Adrenals appeared normal.

KUB

RK : Right Kidney measured 9.5 X 3.9 cms. Cortex and collecting System of right kidney appeared normal. No calculi seen.

LK : Left Kidney measured 9.7 X 4.1 cms. Cortex and collecting

System of left kidney appeared normal. No calculi seen.

BLADDER : Bladder appeared normal.

UTERUS&OVARIES:

Not visualized consistent with post hysterectomy status.

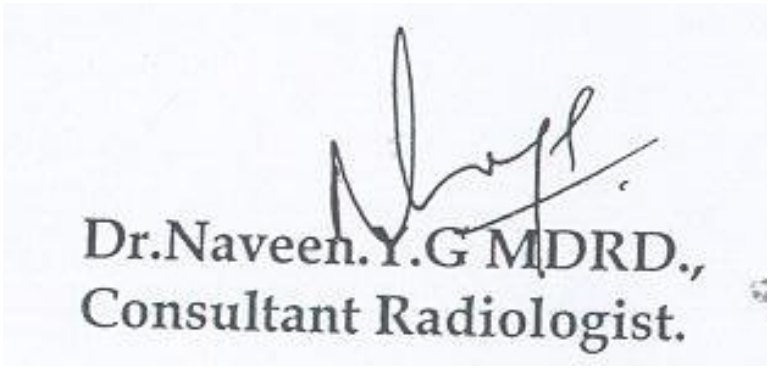
P.O.D:

P.O.D is free.

No adnexal mass lesion seen.

Impression:

Mild hepatomegaly.



AARTHI SCANS

AARTHI SCANS™

AN ISO 9001 ORGANISATION

ABDOMEN & KUB SCAN REPORT

Patient Name:	Mrs. Saraswathy	Age/Sex:	44/f
Referred by:	Dr.Prabhapathy	Visit Date:	10/9/2012

Liver : Liver shows normal. No abscess or mass lesion seen. Liver measured – 12.0cms.

Gall Bladde : Gall Bladder appeared normal. No calculus seen in Gall Bladder. CBD appeared normal. No calculus seen in CBD.

Pancreas : Pancreas appeared normal.

Spleen : Spleen appeared normal. Spleen measured 9.8cms

Aorta : Aorta appeared normal. No Para aortic nodes seen.

Peritoneal cavity: Peritoneal cavity appeared normal.

Adrenals : Adrenals appeared normal.

KUB

RK : Right Kidney measured 9.5 X 3.9 cms. Cortex and collecting System of right kidney appeared normal. No calculi seen.

LK : Left Kidney measured 9.7 X 4.1 cms. Cortex and collecting

System of left kidney appeared normal. No calculi seen.

BLADDER : Bladder appeared normal.

UTERUS&OVARIES:

Not visualized consistent with post hysterectomy status.

P.O.D:

P.O.D is free.

No adnexal mass lesion seen.

Impression:

Normal study.

Dr.Naveen.Y.G MDRD.,
Consultant Radiologist.

MARUTHY HI-TECH
DIAGNOSTIC CENTRE

Govt. Royapettah Hospital Signal, Royapettah, Chennai - 600 014.
Phone : 044 - 28351299, 42105749

Name : Mr. Rajan	Date : 26/10/19
Age : 39 Sex : Male.	Ref By : C.R.I.
Clinical Information :	Consultant's Name : Dr. Arunachalam D.M.R.D

ULTRA SONOGRAM REPORT

USG - ABDOMEN

Liver is enlarged measures 17.0 cms with increased echotexture.

Gall Bladder – Normal. No evidence of calculus

CBD – Normal. No evidence of calculus.

Portal Vein – Normal.

Pancreas – Normal.
Spleen – Normal.

Right Kidney measures 9.9 x 4.0 Cms.
Left Kidney measures 10.0 x 4.1 Cms.
Both kidney shows normal pelvic calyceal system.
No evidence of calculus or calyceal dilatation in both kidneys.

Bladder – Normal.
No evidence of calculus or focal mass lesion.

Prostate – Normal.

Both Illiac fossa normal.

No focal mass or free fluid in the abdomen.

IMPRESSION : HEPATOMEGALY WITH FATTY INFILTRATION OF LIVER
NORMAL KUB

Consultant

Thank you very much for your reference

MARUTHY HI-TECH
DIAGNOSTIC CENTRE
Govt. Royapettah Hospital Signal, Royapettah, Chennai - 600 014.
Phone : 044 - 28351299, 42105749

ABDOMEN & KUB SCAN REPORT

Patient Name:	Mr.Rajan	Age/Sex:	39/M
Referred by:	Dr.Prabhapathy	Visit Date:	4/1/2013

Liver : **Liver shows normal. No abscess or mass lesion seen. Liver measured – 12.0cms**

Gall Bladder : **Gall Bladder appeared normal. No calculus seen in Gall Bladder. CBD appeared normal. No calculus seen in CBD.**

Pancreas : **Pancreas appeared normal.**

Spleen : **Spleen appeared normal. Spleen measured 9.8cms**

Aorta : **Aorta appeared normal. No Para aortic nodes seen.**

Peritoneal cavity: **Peritoneal cavity appeared normal.**

Adrenals : **Adrenals appeared normal.**

KUB

RK : **Right Kidney measured 9.5 X 3.9 cms. Cortex and collecting System of right kidney appeared normal. No calculi seen.**

LK : **Left Kidney measured 9.7 X 4.1 cms. Cortex and collecting System of left kidney appeared normal. No calculi seen.**

BLADDER : **Bladder appeared normal.**

PROSTATE : **Prostrate appeared normal. Measured in 2.8 X 2.5 X 1.9 cms (Weight = 14.4 gms). No intra vesicle enlargement of prostate gland** Normal Study

IMPRESSION

Normal study.

DR.S.NANDINI AZHAR, MBBS.,DMRD.,
RADIOLOGIST

Dr. S. NANDINI, MBBS., DMRD.,
CONSULTANT RADIOLOGIST
REGN. No. 43367

TABLES FOR TRAIL DRUG-2 CHITHIRAMOOLA RASAYANAM

QUALITATIVE ANALYSIS:

Trail drug 2-Table 1

.S.NO	**PARAMETERS**	**RESULTS**
1.	Phosphate	Present
2.	Sulphate	Absent
3.	Magnesium	Absent
4.	Iron	Present
5	Aminoacids	Present
6.	Starch	Absent
7.	Flavonoids	Absent
8.	Proteins	Absent
9.	Tannic acid	Present
10.	Glycosides	Absent

PHYSICAL PROPERTIES

Trail drug 2 table2

S.NO	Characteristic test	Results
1.	pH	4.90
2.	Ash Value	0.98
3.	Water soluble ash	0.02
4.	Acid insoluble ash	0.01

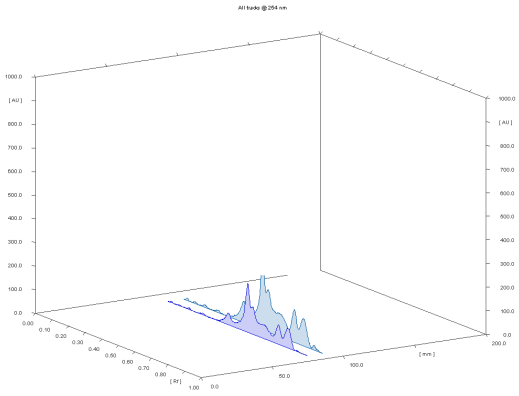
Preliminary acid, basic radicals screening of Chithiramoola Rasayanam.

Trail drug 2 table3

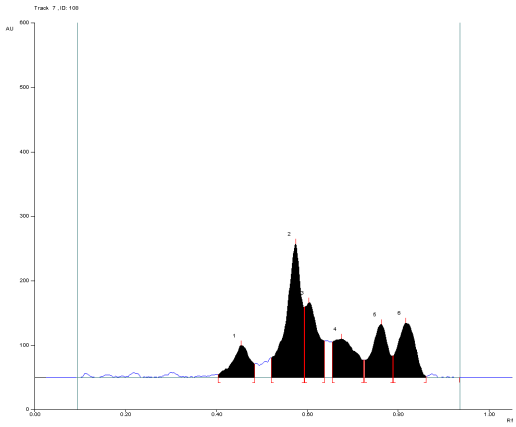
S.No.	Constituents	CMR
1.	Calcium	_
2.	Iron (Ferric)	+
3.	Iron (Ferrous)	+
4.	Chloride	_
5.	Phosphate	+
6.	Potassium	_
7.	Sodium	+
8.	Sulphate	_

108 – HPTLC Profile

254nm

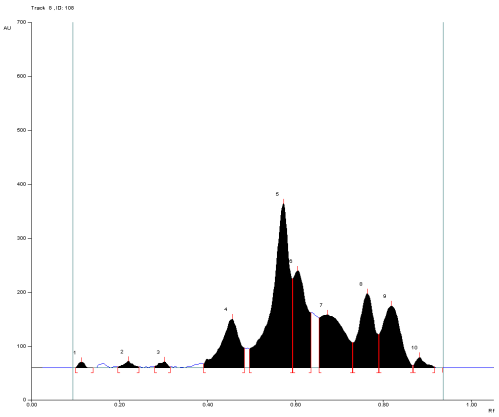


254nm 3D display

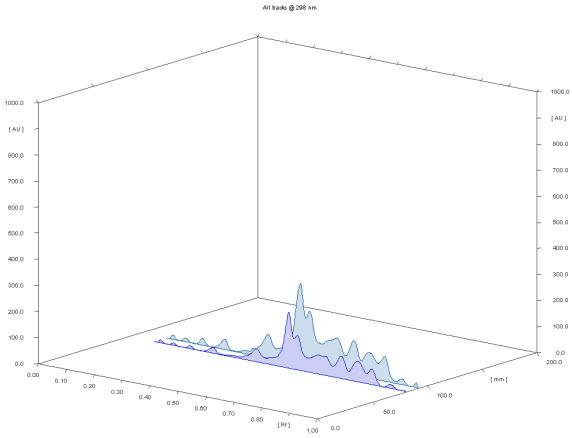


5µl (254nm)

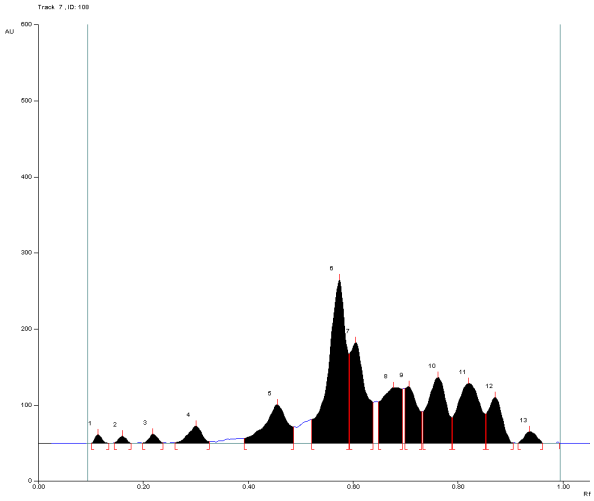
294nm



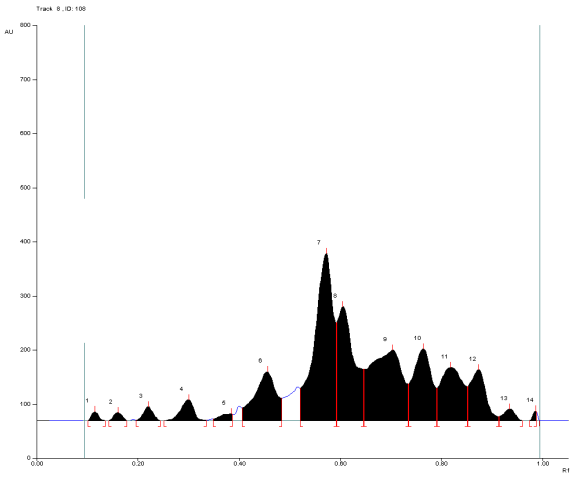
10µl (254nm)



294nm 3D display

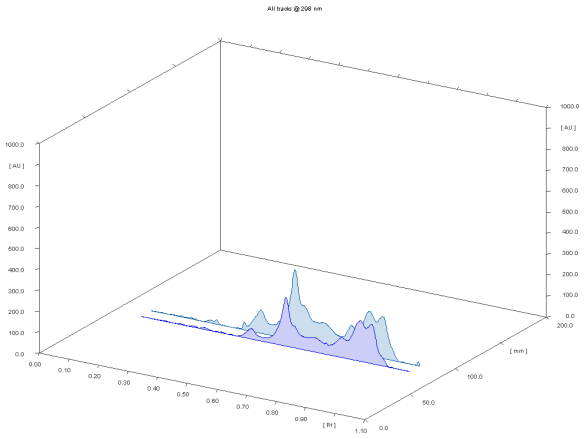


5µl (294nm)

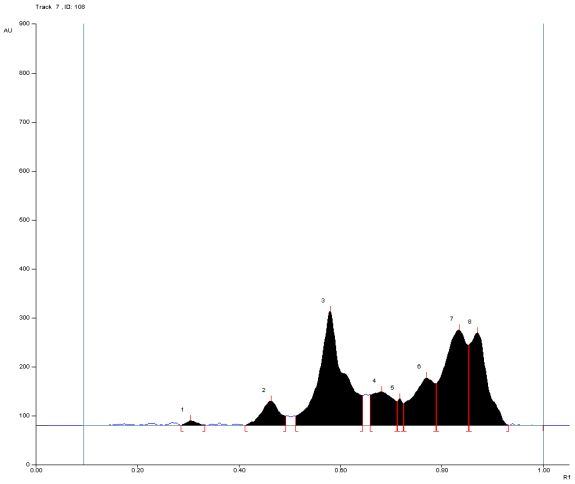


10µl (294nm)

Derivatisation (298nm)

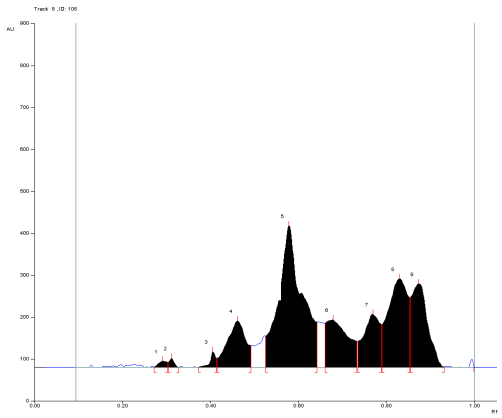


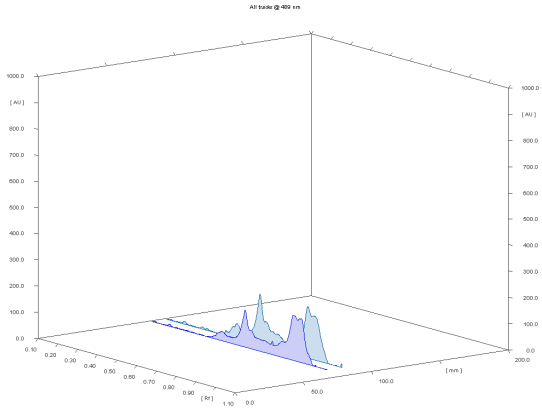
5µl (298nm)



10µl (298nm)

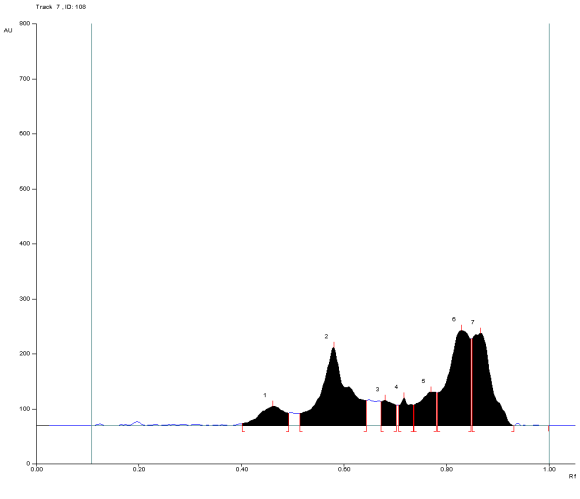
Derivatisation (498nm)

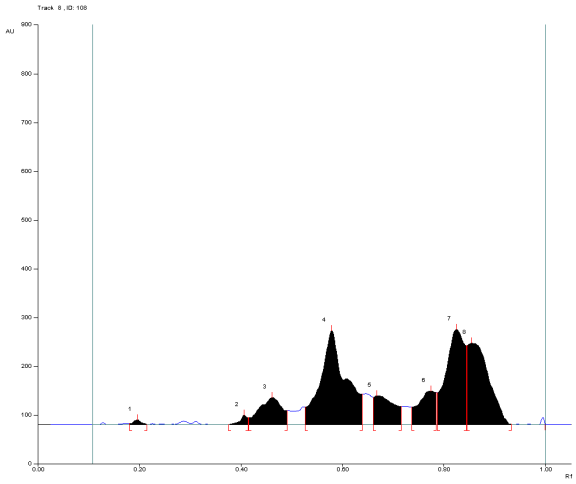




Derivatisation 10µl (298nm)

498 3D display





Derivatisation 5µl (498nm)

Derivatisation 10µl (498nm)

Dose finding experiment and its behavioural Signs of Toxicity

Trail drug 2 table4

No	Dose mg/kg	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
1.	**1000**	+	-	-	+	-	+	-	-	-	-	-	-	-	+	-	-	-	-	-	-
2	**2000**	+	-	-	+	-	+	-	-	-	-	-	-	-	+	-	-	-	-	-	-
3	**5000**	+	-	-	+	-	+	-	-	-	-	-	-	-	+	-	-	+	-	-	-

1. Alertness 2. Aggressiveness 3. Pile erection 4. Grooming 5. Gripping 6. Touch Response 7. Decreased Motor Activity 8. Tremors 9. Convulsions 10. Muscle Spasm 11. Catatonia 12. Muscle relaxant 13. Hypnosis 14. Analgesia 15.Lacrimation 16. Exophthalmos 17. Diarrhoea 18. Writhing 19. Respiration 20. Mortality

Body wt (g) of albino rats exposed to *Chithiramoola Rasayanam* for 28days.

Trail drug 2 table5

Dose (mg/kg/day)	Days				
	1	**7**	**14**	**21**	**28**
Control	164.06±1.80	165.51±1.10	167.60±2.5	168.75±2.7	168.75±2.2
100	150.1±1.22*	151.6±1.82*	152.51±0.20**	154.50±0.2	155.3±0.3
250	152.4±0.72	153.14±1.04*	155.00±1.32*	160.25±2.36	160.20±2.3
500	145.08±7.4**	146.22±6.20**	153.55±4.91**	148.61±11.2	149.02±7.10**

Values are mean ± S.E.M. (Dunnett's test). *P<0.05; **P<0.01. N=6.

Food (g/day) intake of rats exposed to *Chithiramoola Rasayanam* for 28days.

Trail drug 2 table6

Dose (mg/kg/day)	Days (gms/rats)				
	1	**7**	**14**	**21**	**28**
Control	41.18±2.54	40.12±2.15	42.17±2.16	45.11±2.14	42.18±2.17
100	40.45±2.74	45.15±2.14	44.48±2.24	45.00±2.91	42.10±2.48
250	40.24±2.47	42.33±2.10	42.84±2.45	44.26±2.20	43.24±2.15
500	41.18±2.26	40.48±2.28	43.28±2.48	40.10±2.22	42.34±2.45

Values are mean ± S.E.M. (Dunnet 't' test). $^{ns}P>0.05$ Vs Control N=6.

Water (ml/day) intake of rats exposed to *Chithiramoola Rasayanam* for 28days.

Trail drug 2 table7

Dose (mg/kg/day)	Days(ml/rat)				
	1	**7**	**14**	**21**	**28**
Control	45.34±2.31	42.33±2.74	44.16±4.15	45.20±3.35	42.92±3.00
100	40.40±2.27	43.40±3.11	45.66±3.63	44.31±3.00	45.54±2.51
250	42.48±2.72	42.12±2.54	43.12±3.16	42.52±2.64	47.40±3.20
500	46.32±2.61	44.30±2.12	45.90±3.07	45.27±3.44	45.28±3.22

Values are mean ± S.E.M. (Dunnet 't' test). $^{ns}P>0.05$ Vs Control N=6.

Hematological parameters after 28days treatment with *Chithiramoola Rasayanam* in rats.

Trail drug 2 table8

Parameter	Control	100 mg/kg	250 mg/kg	500 mg/kg
Red blood cell (mm^3)	4.45±0.41	4.90±0.43	4.72±0.32	4.28±0.18
HB (%)	14.51±0.45	14.2±0.80	14.52±0.40	14.80±0.28
Leukocyte ($x10^3$/Cu.mm)	3.15±0.4	3.18±0.5	2.60±0.30	2.72±0.37
Platelets(K/µl)	1.12±0.12	1.42±0.25	1.42±0.25	1.52±0.22
MCV (gl)	85.13±4.4	86.12±4.10	85.51±3.20	85.12±7.02
N	50.10±4.50	45.10 ±2.7	47.22±2.8	47.20±3.2
L	47.00±0.6	46.14±3.18	48.24±1.42	50.60±2.01
M	4.0±0.02	3.4±0.2*	4.0±0.3	3.2±0.2*
E	6.2±0.40	5±0.5	5±0.4	5±0.4
B	0±0.00	0±0.00	0±0.00	0±0.00
ESR(mm)	1±00	1±00	1±00	1±00
PCV	44.12±2.0	43.21±2.30	42.10±2.44	42.20±3.02

Effect of treatment with *Chithiramoola Rasayanam* biochemical parameters.

Trail drug 2 table9

Dose (mg/kg)	Control	100 mg/kg	250 mg/kg	500 mg/kg
Total Bilirubin (mg/dL)	0.23±0.01	0.24±0.03	0.25±0.03	1.41±1.07
Bilirubin direct (mg/dL)	0.00±0.00	0.02±0.01	0.00±0.00	0.13±0.13
ALP (U/L)	51.20±4.25	38.80±2.80	53.60±3.28	50.60±6.29
SGOT (U/L)	125.2±6.70	130.10±5.29	137.90±7.16	215.10±62.70
SGPT(U/L)	24.52±1.40	25.32±2.12	27.00±1.80	47.12±3.58**
Total Protein(g/dl)	5.12±0.26	5.54±0.21	5.52±0.14	6.10±0.30*
Albumin(g/dl)	3.90±0.06	3.71±0.05	3.62±0.07*	3.73±0.07
Globulin(g/dl)	3.98±0.15	4.10±0.22	4.08±0.16	4.56±0.24

Values are mean ± S.E.M. *P<0.05; **P<0.01. Vs Control N=6.

Trail drug 2 table10

Dose (mg/kg)	Control	100 mg/kg	250 mg/kg	500 mg/kg
Urea(µg/dL)	30.67±4.3	37.67±4.3	38.00±5.6	34.50±4.68
Creatinine (mg/dL)	29.38±2.2	31.38±3.3	28.01±2.01	29.19 ± 5.2
Uric acid (mg/dL)	3.30±0.05	1.98±0.07**	2.10±0.08**	1.37±0.08**
Na m.mol	149.80±0.84	148.00±1.00	147.20±1.10	146.60±0.55
K m.mol	6.12±0.58	5.90±1.00	5.68±0.08	5.82±0.08
Cl m.mol	98.14±3.54	102.78±5.00	104.35±4.95	102.70±5.44

Values are mean ± S.E.M. **P<0.01. Vs Control N=6.

Lipid Profile

Trail drug 2 table11

Dose (mg/kg)	Control	100 mg/kg	250 mg/kg	500 mg/kg
Total cholesterol (mg/dL)	56.86±5.77	57.61±5.68	55.84±5.21	52.50±3.04
HDL(mg/dL)	95.31±0.25	104.5±0.22**	163.67±0.27**	150.39±0.49**
LDL(mg/dL)	81.44±2.50	32.48±4.75**	95.60±0.60*	71.77±3.50
VLDL(mg/dl)	25.10±2.00	24.72±2.24	25.48±2.95	25.24±2.48
Triglycerides (mg/dl)	25.3±4.2	27.33±5.2	28.94±4	30.75±4.2
Blood glucose(mg/dl)	102±13.4	90±10.6	105±18.7	113±15.52

*Values are mean ± S.E.M. *P<0.05; **P<0.01. Vs Control N=6.*

Urine Analysis

Trail drug 2 table12

Parameters	**Control**	**100 mg/kg**	**250 mg/kg**	**500 mg/kg**
Colour	Yellow	Yellow	Yellow	Yellow
Transparency	Clear	Slightly turbid	Slightly cloudy	Slightly turbid
Specific gravity	1.010	1.010	1.010	1.010
PH	>7.2	>8.0	>7.5	>7.5
Protein	Nil	1+	1+	2+
Glucose	Nil	Nil	Nil	Trace
Bilirubin	-ve	-ve	-ve	-ve
Ketones	-ve	-ve	-ve	-ve
Blood	Absent	Absent	Absent	Absent
Urobilinogen	Normal	Normal	Normal	Normal
Pus cells	0-cells/HPF	1-cell/HPF	2-cells/HPF	1-cell/HPF
RBCs	Nil	Nil	0-1cells/HPF	Nil
Epithelial cells	Nil	1-cell/HPF	Nil	1-cell/HPF
Crystals	Nil	Nil	Nil	Nil
Casts	Nil	Nil	Nil	Nil
Others	Bacteria seen	Bacteria seen	Bacteria seen	Bacteria seen

Effect of oral administration of *Chithiramoola Rasayanam* on organ weight

Trail drug 2 table13

Dose (mg/kg)	Control	100 mg/kg	250 mg/kg	500 mg/kg
Liver (g)	6.50±0.44	6.72±0.38	6.44±0.32	6.35±0.30
Heart (g)	1.24±0.05	1.31±0.05	1.17±0.04	1.25±0.05
Lung (g)	1.88±0.13	1.72±0.05	1.78±0.12	1.68±0.06
Spleen (g)	0.90±0.04	0.85±0.03	0.77±0.03*	0.81±0.04
Ovary (g)	0.07±0.00	0.08±0.00	0.09±0.00	0.06±0.03
Testes (g)	2.00±0.10	2.15±0.15	2.24±0.20	2.05±0.18
Brain (g)	1.90±0.02	1.91±0.03	1.80±0.03*	1.92±0.03
Kidney (g)	1.26±0.03	1.25±0.03	1.14±0.02*	1.25±0.03
Stomach (g)	1.25±0.18	1.20±0.12	1.14±0.14	1.18±0.15

Values are mean ± S.E.M. (Dunnet 't' test). *P<0.05; Vs Control N=6.

Analgesic activity of Chitramoola Rasayanam in mice

Trail drug 2 table14

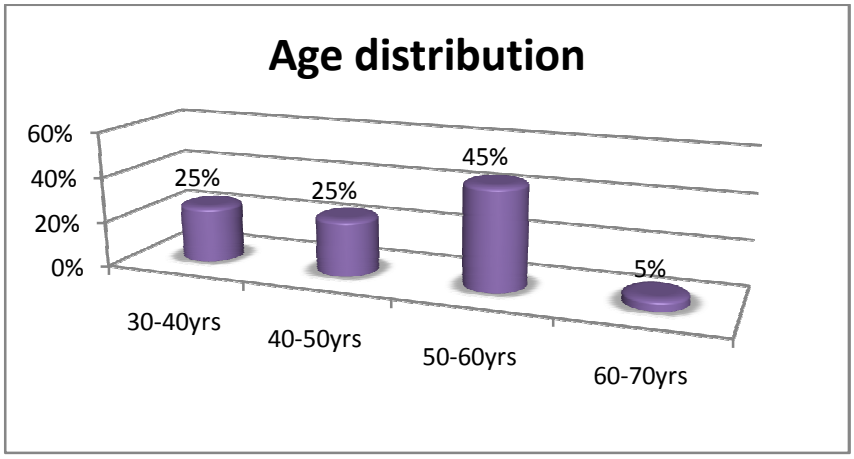
Drug and Route	Dose (mg/kg)	No. of animals	Reaction time before treatment	% increase in Reaction time in Sec after CMR treatment (Mean ± SEM)				
				15min**	30min**	45min**	60min**	90min**
Control	saline	6	3.0±0.05	5.2±0.8	12.4±0.5	12.1±0.04	13.5±0.6	14.16±0.4
CMR	100	6	3.1±0.04	8.2±0.12	13.8±0.10	24.5±0.44	31.18±1.20	34.12±2.18
CMR	250	6	2.9±0.05	10.1±0.12	20.1±0.14	35.6±0.52	46.20±1.15	49.00±2.32
CMR	500	6	3.0±0.08	11.6±0.10	31.4±0.12	41.2±0.5	48.89±1.24	52.10±2.00
Pentazocine	5	6	3.3±0.14	31.10±0.24	52.1±0.32	70.02±1.03	69.38±2.70	68.13±1.77

Values are expressed as mean±SEM; ** $P<0.01$, as compared to their control

CLINICAL ASSESMENT:

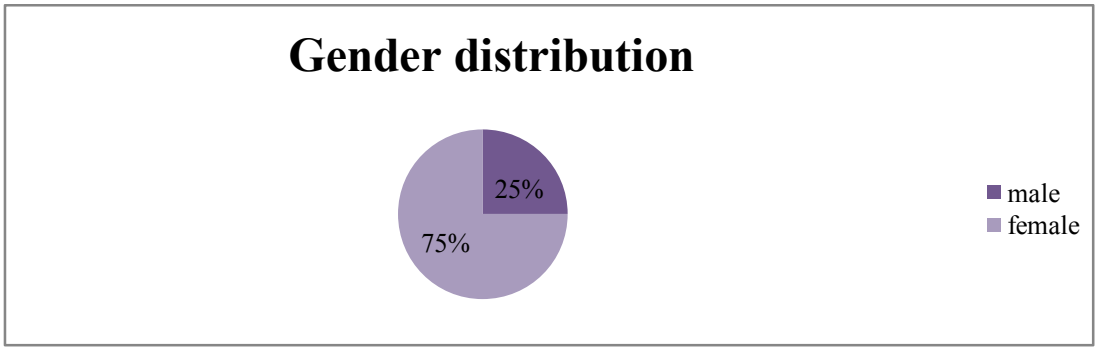
AGE DISTRIBUTION

AGE YRS	NO OF PATIENT	%
30-40	5	25
40-50	5	25
50-60	9	45
60-70	1	5



GENDER DISTRIBUTION

S.NO	GENDER	NO.OF PATIENTS	PERCENTAGE
1.	**FEMALE**	**5**	**25%**
2.	**MALE**	**15**	**75%**

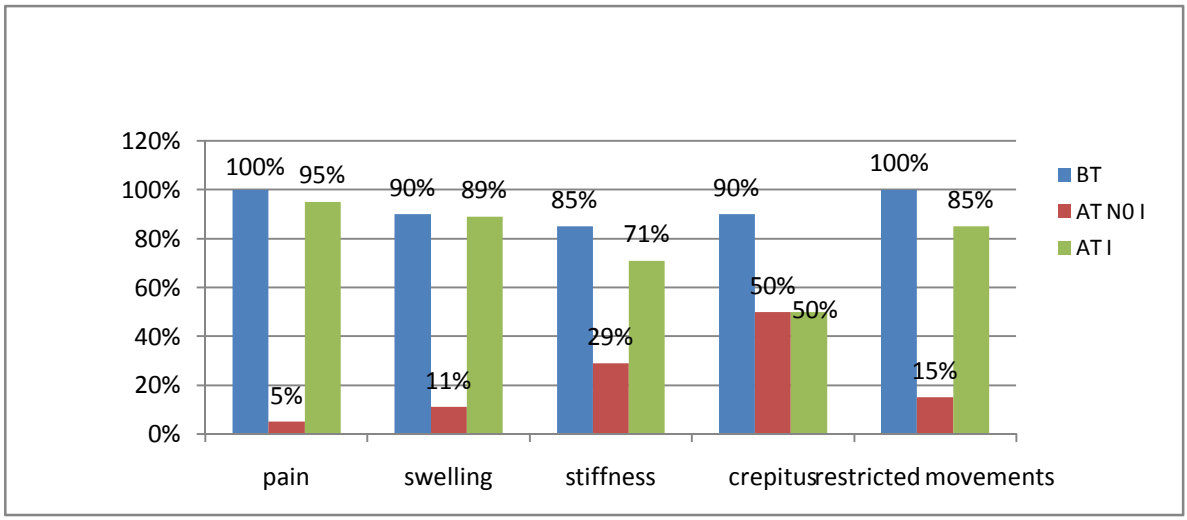


IMPROVEMENT IN THE PAIN SCORE OF THE SOOLAI PATIENTS

S.NO	OPD NO/ IP NO.	AGE	BT	AT
1	4934	58	7	4
2	C83793	40	6	2
3	C24806	62	4	1
4	C85159	39	6	2
5	C87411	49	5	2
6	4972	39	7	3
7	4018	43	3	1
8	4989	34	6	0
9	C87560	50	3	0
10	C84893	50	4	1
11	C89473	56	5	2
12	4060	56	6	1
13	C90735	50	4	2
14	4124	49	5	2
15	C89474	52	7	3
16	5068	55	6	2
17	C86851	52	4	0
18	C86535	36	5	2
19	C94410	35	4	1
20	C95642	44	4	2

IMPROVEMENT SHOWING SIGNS AND SYMPTOMS BEFORE AND AFTER TREATMENT OF SOOLAI .

SYMPTOMS	NO. OF PATIENTS WITH SYMPTOMS			
	BT	AFTER TREATMENT		IMPROVEMENT PERCENTAGE
		NO IMPROVEMENT	IMPROVEMENT	
PAIN	20 (100%)	1(5%)	19 (95%)	95%
SWELLING	18(90%)	2(11%)	16 (89%)	89%
STIFFNESS	17(85%)	5(29%)	12 (71%)	71%
CREPITUS	18(90%)	9(50%)	9 (50%)	50%
RESTRICTED MOVEMENTS	20(100%)	3(15%)	17 (85%)	85%



BT-BEFORE TREATMENT

AT- AFTER TREATMENT

AT - AFTER TREATMENT NO IMPROVEMENT

IMPROVEMENT IN PROGNOSIS OF SYMPTOMS OF SOOLAI.

S.N0	op/ip	AGE	SEX	BTKP	ATKP	BTKS	ATKS	BTSTIFF	ATSTIFF	BTCRE	ATCRE	BTRES	ATRES	BTSYM	ATSYM
1	4934	58	M	+	_	_	_	+	_	+	+	+	_	4	1
2	C83793	40	F	+	_	+	_	+	_	+	_	+	_	5	0
3	3983	62	F	+	_	+	_	+	+	+	+	+	+	5	4
4	C85159	39	F	+	_	+	_	_	_	+	+	+	_	4	1
5	C87411	49	F	+	+	+	_	+	_	+	+	+	+	5	3
6	4972	39	M	+	_	+	_	_	_	+	_	+	_	3	0
7	4018	43	F	+	_	+	_	+	_	+	+	+	_	5	1
8	4989	34	M	+	_	+	_	_	_	+	_	+	_	4	0
9	C87560	50	F	+	_	+	_	+	_	+	+	+	_	5	1
10	C84893	50	M	+	_	+	_	+	+	+	+	+	_	5	1
11	C89473	56	F	+	_	_	_	+	_	+	+	+	_	4	1
12	4060	56	F	+	_	_	_	+	_	+	+	+	_	4	1
13	C90735	50	F	+	_	+	_	+	_	+	_	+	_	5	0
14	4124	49	F	+	_	+	_	+	_	+	+	+	+	5	3
15	C89474	52	F	+	_	+	_	+	+	+	+	+	_	5	2
16	5068	55	M	+	_	+	_	+	_	+	_	+	_	5	0
17	C86851	52	F	+	_	+	_	+	_	+	+	+	_	5	1
18	C86535	36	F	+	_	+	_	+	_	_	+	+	_	5	1
19	C94410	27	F	+	_	+	_	+	+	+	_	+	_	5	1
20	C95642	44	F	+	_	+	_	+	+	_	_	+	_	5	1

BT-BEFORE TREATMENT,KP-KNEE PAIN, KS-KNEE SWELLING ,STIFF- STIFFNESS,CRE-CREPITUS,RES-RESTRICTION, SYM-SYMPTOMS.
AT-AFTER TREATMEN

SPECIFIC INVESTIGATION FOR SOOLAI

S.NO	OP/IP	AGE	SEX	BTHB Mg/dl	ATHB Mg/dl	BTHESR MM	ATHESR MM	BTOESR MM	ATOESR MM	BTDCP %	ATDCP %	BTDCL %	ATDCL %	BTSCAL Mg/dl	ATSCAI Mg/dl
1	4934	58	M	12	12.2	4	2	8	4	58	54	25	39	12	11.6
2	C83793	40	F	12	12.7	30	4	64	20	63	64	34	30	10	9.8
3	C24806	62	F	13	12.8	42	6	86	20	86	20	36	35	10	10.9
4	C85159	39	F	14	13.1	2	4	8	16	30	69	68	26	11	10
5	C87411	49	F	13	12.6	4	8	20	40	55	65	40	30	11	9.8
6	4972	39	M	15	14.9	2	2	4	10	64	65	33	31	11	11
7	4018	43	F	14	13.8	4	4	8	16	50	50	46	47	10	10
8	4989	34	M	18	15.6	2	2	4	4	54	50	40	37	11	10.7
9	C87560	50	F	12	11.9	2	2	4	6	60	62	31	33	10	10.6
10	C84893	50	M	15	14.9	2	2	4	4	68	60	28	30	12	10.2
11	C89473	56	F	13	12.7	6	4	20	14	53	50	41	40	11	10.5
12	4060	56	F	13	13.6	8	4	16	5	61	50	36	45	11	10
13	C90735	50	F	13	13	4	2	10	6	65	60	34	29	11	11.2
14	4124	49	F	14	11.9	8	4	20	12	59	50	35	46	11	10
15	C89474	52	F	13	12.3	10	2	22	6	72	67	26	29	12	10.6
16	5068	55	M	14	10.9	2	2	4	8	65	62	35	28	11	10.6
17	C86851	52	F	13	11.9	8	2	16	8	59	62	35	34	11	10
18	C86535	36	F	13	11	4	2	8	4	60	60	35	36	9.9	10.8
19	C94410	27	F	12	10.8	2	2	8	10	56	55	39	40	10	10.5
20	C95642	44	F	14	11.4	10	6	24	12	65	67	34	30	11	10.3

Drug2 BEFORE TREATMENT INVESTIGATION

S.NO	OP/IP	AGE	SEX	HB Mg/dl	TC Cells/cu. mm	DC				FBS Mg/d	PPBS mg/dl	UREA mg/dl	CREm mg/dl	URI mg/dl	CHO mg/dl	HDL mg/dl	LDL mg/dl	VLDL mg/dl	TB Mg/dl	DB Mg/dl	IB Mg/dl	OT U/L	PT U/L	ALP Mg/dl	TP Mg/dl	ALB Mg/dl	GLO Mg/dl	ASO	CRP	RAF	UALB	UFS	UPS	P.C	E.P
						P	L	M	N																									
1	4934	58	M	12.2	7700	58	25	17	0	110	112	18	0.6	6.9	237	37	147	77	O.7	0.5	0.2	23	25	149	7	5	2	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
2	C83793	40	F	12.3	11300	63	34	3	0	95	134	17	0.5	6	190	30	121	40	0.7	0.2	0.5	11	12	162	6	4	2	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
3	C24806	62	F	13	6200	57	36	6	1	95	103	14	0.5	3.8	142	38	64	22	0.5	0.3	0.2	29	30	196	7.6	4.2	3.4	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
4	C85159	39	F	13.6	8700	30	68	2	0	89	124	23	0.6	4.5	138	24	78	29	0.7	0.3	0.4	20	21	182	6	4	2	NEG	NEG	NEG	NIL	NIL	NIL	I-2P	I-2E
5	C87411	49	F	12.9	8000	55	40	5	0	110	160	14	0.4	5.8	232	40	50	67	0.6	0.2	0.4	22	24	164	7.4	4.2	3.2	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
6	4972	39	M	14.9	6700	64	33	3	0	91	134	16	0.5	4	173	32	93	58	0.5	0.2	0.3	21	24	217	6.7	4.7	2	NEG	NEG	NEG	NIL	NIL	NIL	2-4P	2-4E
7	4018	43	F	13.6	6300	50	46	4	0	84	108	28	0.7	5.6	180	30	100	40	0.7	0.3	0.2	26	27	170	7.4	4	3.7	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
8	4989	34	M	17.7	9200	54	40	6	0	95	105	22	0.6	7.3	151	32	62	31	0.5	0.2	0.3	25	26	179	6.6	4	2.6	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
9	C87560	50	F	11.6	9600	60	31	9	0	101	139	20	0.6	4.5	158	39	70	42	0.4	0.2	0.2	14	16	130	7	5	2	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
10	C84893	50	M	15.3	7700	68	28	4	0	109	132	24	0.6	4.8	191	35	132	18	0.6	0.2	0.4	18	19	130	7	4.2	2.8	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
11	C89473	56	F	13.1	9700	53	41	6	0	118	122	16	0.5	6.6	201	35	101	76	0.3	0.2	0.1	12	11	132	7.6	5.2	2.4	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
12	4060	56	F	13.4	7900	61	36	3	0	105	124	27	0.7	3.8	182	40	126	18	0.5	0.2	0.3	14	16	162	6.6	4.1	2.5	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
13	C90735	50	F	12.6	6400	65	34	1	0	99	118	16	0.5	4.1	170	36	123	19	0.6	0.2	0.4	20	21	150	6.6	4.2	2.4	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
14	4124	49	F	14.1	7200	59	35	6	0	89	119	22	0.6	4.5	210	40	126	45	0.4	0.2	0.2	12	14	154	7	4.6	2.4	NEG	NEG	NEG	NIL	NIL	NIL	2-4P	2-4E
15	C89474	52	F	12.5	10300	72	26	2	0	90	132	14	0.4	5.5	216	36	140	34	0.5	0.2	0.3	24	25	198	7.9	5.3	2.6	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
16	5068	55	M	13.1	7100	65	35	10	0	100	120	27	0.6	4	206	36	126	27	0.6	0.2	0.4	26	29	243	6	4	2	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
17	C86851	52	F	12.6	6300	59	35	5	0	112	133	20	0.7	3.3	193	40	26	18	0.5	0.2	0.3	13	15	152	6.2	4.2	2	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
18	C86535	36	F	12.7	9000	60	35	5	0	84	132	26	0.7	4.4	128	30	92	17	0.6	0.2	0.4	23	25	149	6.7	4.2	2.5	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
19	C94410	35	F	11.8	4800	56	39	5	0	103	115	15	0.4	5	135	30	101	20	0.5	0.2	0.3	14	16	172	6.4	4.1	2.3	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
20	C95642	44	F	13.6	7600	65	34	1	0	101	118	17	0.5	4	171	35	121	24	0.6	0.2	0.4	14	18	226	7.1	4.4	2.7	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E

AFTER TREATMENT –SOOLAI

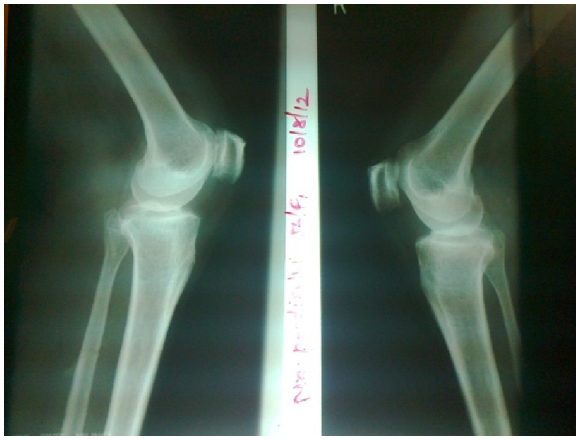
S. NO	OP/IP	AGE	SEX	ATHB Mg/dl	ATTC	ATDC				FAS Mg/dl	PPAS mg/dl	UREA mg/dl	CRE mg/dl	URIC mg/dl	CHO mg/dl	HDL mg/dl	LDL mg/dl L	VLDL mg/dl	TB mg/dl	DB mg/dl	IB mg/dl	OT u/l	PT u/l	ALP mg/dl	TP mg/dl	ALB mg/dl	GLO mg/dl	ASO	CRP	RAF	UALB	UFS	UPS	PUS CELLS	EPI.CELLS
						P	L	E	m																									
1	4934	58	M	12.3	7700	54	39	7	0	110	120	18	0.5	3.1	200	43	120	40	0.4	0.2	0.2	25	25	120	7	5	2	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
2	C83793	40	F	12.7	11600	64	30	6	0	94	104	17	0.5	3	205	40	125	45	0.7	0.3	0.4	12	15	164	6	4	2	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
3	C24806	62	F	12.8	6200	40	35	24	1	108	121	18	0.5	3.6	141	32	106	21	0.7	0.3	0.4	17	18	159	5.9	3.2	2.7	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
4	C85159	39	F	13.1	9000	69	26	5	0	79	103	15	0.4	3.2	115	30	60	23	0.5	0.2	0.3	19	21	150	5.7	3.7	2	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
5	C87411	49	F	12.3	9500	65	30	5	0	110	130	21	0.6	4.7	246	49	132	77	0.5	0.2	0.3	16	18	151	7	5	2	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
6	4972	39	M	14.9	8900	65	31	4	0	118	137	19	0.5	4	165	40	72	35	0.5	0.2	0.3	17	18	156	5.8	3.1	2.7	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
7	4018	43	F	13.8	6500	50	47	3	0	92	134	22	0.6	3.8	167	36	107	23	0.6	0.2	0.4	12	12	157	7	5	2	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
8	4989	34	M	15.6	9400	50	37	13	0	100	133	20	0.6	4.8	115	26	26	21	0.5	0.2	0.3	12	14	201	5	3	2	NEG	NEG	NEG	NIL	NIL	NIL	2-3P	2-3E
9	C87560	50	F	11.9	11900	62	33	5	0	105	120	20	0.6	□ 1	140	35	86	28	0.5	0.2	0.3	16	17	189	5	3	2	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
10	C84893	50	M	14.9	7500	60	30	5	0	88	100	18	0.5	3	200	40	99	26	0.7	3	0.4	17	19	175	6.2	3.7	2.5	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
11	C89473	56	F	12.7	7800	50	40	10	0	100	110	15	0.4	4.3	200	40	48	98	0.4	0.2	0.2	14	15	159	5.1	3	2.1	NEG	NEG	NEG	NIL	NIL	NIL	4-5P	4-5E
12	4060	56	F	13.6	6700	50	45	5	0	91	102	15	0.6	3.3	177	36	80	28	0.7	0.3	0.4	17	19	159	6.5	3.6	2.9	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
13	C90735	50	F	13	5800	60	29	11	0	92	106	20	0.9	3.3	173	45	86	35	0.5	0.2	0.3	12	14	196	5.7	3.1	2.6	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
14	4124	49	F	11.9	6800	50	46	4	0	107	125	14	0.7	4.1	239	47	119	91	0.5	0.2	0.3	17	19	179	6.2	3.9	2.3	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
15	C89474	52	F	12.3	9800	67	29	4	0	117	124	26	0.7	3.4	200	42	132	46	0.4	0.2	0.2	16	18	120	6.3	3.7	2.6	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
16	5068	55	M	10.9	8200	62	28	10	0	102	137	20	0.7	3.9	200	34	101	13	0.5	0.2	0.3	24	26	216	6.9	3.8	3.1	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
17	C86851	52	F	11.9	5900	62	34	4	0	127	157	23	0.7	3.4	210	33	108	26	0.5	0.2	0.3	20	22	183	6.9	3.7	3.2	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
18	C86535	36	F	11	6100	60	36	4	0	95	107	15	0.5	3	131	31	69	10	0.6	0.2	0.4	11	12	152	5.2	3.1	2.1	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
19	C94410	35	F	11	4800	55	40	5	0	113	123	14	0.5	3	135	33	72	87	0.7	0.3	0.4	15	17	159	6.7	4.2	2.5	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E
20	C95642	44	F	11.4	9500	67	30	3	0	94	104	16	0.6	170	33	78	12	64	0.6	0.2	0.4	12	13	181	3.7	2.3	1.4	NEG	NEG	NEG	NIL	NIL	NIL	1-2P	1-2E

KNEE JOINT-AP VIEW

BEFORE TREATMENT

AFTER TREATMENT

KNEE JOINT (LATERAL VIEW)



BEFORE TREATMENT

AFTER TREATMENT

STATISTICAL ANALYSIS:

All collected data were entered into MS Excel software using different columns as variables and rows as patients. SPSS software was used to perform statistical analysis. Basic descriptive statistics include frequency distributions and cross-tabulations were performed. The quantity variables were expressed as Mean ± Standard Deviation and qualitative data as percentage. A probability value of <0.05 was considered to indicate as statistical significance. Paired 't' test was performed for determining the significance between before and after treatment.

DRUG 2

SOOLAI

Paired t test for Symptoms before and after treatment:

Variable	**Obs**	**Mean**	**Std.dev**	**t.value**	**P value**
Before treatment	20	4.65	0.587	14.888	P<0.0005
After treatment	20	1.15	1.089		

Symptoms before treatment is 4.65 and after treatment is 1.089 which is statistically significant($p<0.0005$).

CERTIFICATE

This is to certify that the project title: Preclinical study on "Chitramoola Rasayanam" for Anti-Inflammatory Activity in the management of Soolai (Osteoarthritis) has been approved by the IAEC with the reference number XIII/VELS/PCOL/32/2000/CPCSEA/IAEC/08.08.12

Name of Member Secretary IAEC:

Dr. J. Anbu

Name of CPCSEA nominee:

Dr. K. Sadhasivan Pillai

Signature with date

J. Anbu 08/08/12

Member Secretary of IAEC

Dr. J.ANBU, M.Pharm., Ph.D., D.M.L.T.,MBA
Professor & Head
Department of Pharmacology & Toxicology
School of Pharmaceutical Sciences
Vels University
Pallavaram, Chennai-600 117

CERTIFICATE

This is to certify that the project title: Preclinical study on "Palagarai Parpam" for Hepatoprotective Activity in the management of kalleral noi" has been approved by the IAEC with the reference number XIII/VELS/PCOL/33/2000/CPCSEA/IAEC/08.08.12

Name of Member Secretary IAEC:

Dr. J. Anbu

Name of CPCSEA nominee:

Dr. K. Sadhasivan Pillai

Signature with date

08/08/12

Member Secretary of IAEC

Dr. J.ANBU, M.Pharm., Ph.D., D.M.L.T.,MBA
Professor & Head
Department of Pharmacology & Toxicology
School of Pharmaceutical Sciences
Vels University
Pallavaram, Chennai-600 117

The Tamil Nadu Dr. M.G.R. Medical University

69, Anna Salai, Guindy, Chennai-600 032

This Certificate is awarded to ~~Mr/Ms~~/Dr.....K. PRABHAPATHY.....

for participating as a ~~Resource Person~~ / Delegate in the VII Workshop

on **"Research Methodology & Biostatistics"**

for AYUSH Post-Graduates & Researchers

organized by the Department of Siddha

The Tamil Nadu Dr. M.G.R. Medical University

from 6th Feb. 2012 to 10th Feb. 2012.

DR. MAYILVAHANAN NATARAJAN
M.S.Orth. M.Ch.Orth. (L'pool) Ph.D. (Orth. Onco.) F.R.C.S. (Eng) D.Sc.
7th VICE CHANCELLOR

Dr. R. SRILAKSHMI, DCH, Ph.D.
REGISTRAR

Dr. N. KABILAN, M.D. (Siddha)
READER, DEPT. OF SIDDHA

NATIONAL INSTITUTE OF SIDDHA

(An Autonomous Body under Department of AYUSH)
Ministry Of Health & Family Welfare, Government of India

Tambaram Sanatorium, Chennai - 600 047
Tel : 044-22411611 Fax : 044-22381314
E-mail : nischennaisiddha@yahoo.co.in
Website : www.nischennai.org

Name: Dr. K. Prabhapathy Reg. NO 32101703
Title: Preclinical and clinical Study on "Palagarai Parpam for hepato Protective Activity" in the management of Kalleral Noi (Liver disease)
No. NIS/IEC/2011/3/11a - 24/12/2011

DECSION

Opinion of the Institutional Ethics Committee – Please Check one
✓ Approval _____ Modifications required prior to approval (Please specify one space below) _____ Disapproval Date of review: _____ K. Manickavasakam (Dr. K. MANICKAVASAKAM) Member secretary Signed: _____ (Please print name) Dr.V.SUBRAMANIAN chair person (Please delee as appropriate, Chairperson, Secretary)
Modifications needed Modification given to candidate
The research proponent is hereby informed that the Institutional Ethics Committee will require the following: 1. All adverse drug reactions (ADRs) that are both serious and unexpected to be reported promptly to the IEC within 7 working days 2. The progress report to be submitted to the IEC atleast annually 3. Upon completion of the study, a final study status report needs to submitted to the IEC

IAEC Protocol nos 1248/ac/09/CPCSEA/4-11A/2011

20/12/2011

CERTIFICATE

This is certify that the project title. Preclinical and clinical Study on "PALAGARAI PARPAM" for HEPATOPROTECTIVE activity in the management of Kalleral noi (liver disease) **has been approved by the IAEC.**

Prof. Dr. K. Manickavasakam — **Name of Chairman/Member Secretary IAEC:**

Dr. B. Jayachandran Dare — **Name of CPCSEA nominee:**

Signature with date

Chairman/Member Secretary of IAEC:

CPCSEA nominee:

(Kindly make sure that minutes of the meeting duly signed by all the participants are maintained by Office)

NATIONAL INSTITUTE OF SIDDHA

(An Autonomous Body under Department of AYUSH)
Ministry Of Health & Family Welfare, Government of India

Tambaram Sanatorium, Chennai - 600 047
Tel : 044-22411611 Fax : 044-22381314
E-mail : nischennaisiddha@yahoo.co.in
Website : www.nischennai.org

Name: Dr. K. Prabhapathy Reg. No 32101703
Title: Preclinical and Clinical study On "Chitramoola Rasayanam" for "Anti inflammatory Activity" in the management Of Soolai (Osteoarthritis)
No. NIS/IEC/2011/3/116 - 24/12/2011

DECSION

Opinion of the Institutional Ethics Committee – Please Check one
✓ Approval _____ Modifications required prior to approval (Please specify one space below) _____ Disapproval Date of review: _____ K. Manickavasakam (Dr. K. MANICKAVASAKAM) Member Secretary Signed: _____ (Please print name) Dr.V.SUBRAMANIAN chair person (Please delee as appropriate, Chairperson, Secretary)
Modifications needed Modification given to candidate
The research proponent is hereby informed that the Institutional Ethics Committee will require the following: 1. All adverse drug reactions (ADRs) that are both serious and unexpected to be reported promptly to the IEC within 7 working days 2. The progress report to be submitted to the IEC atleast annually 3. Upon completion of the study, a final study status report needs to submitted to the IEC

IAEC Protocol No 1248/ac/09/CPCSEA/4-11B/2011

20/12/2011

CERTIFICATE

This is certify that the project title Preclinical and clinical Study on "CHITHIRA MOOLARASAYANAM" for ANTI-INFLAMMATORY activity in the management OF SOOLAI (Osteoarthritis)

has been approved by the IAEC.

Prof. Dr. K. Manickavasakam — **Name of Chairman/Member Secretary IAEC:**

Dr. B. Jayachandran Dare — **Name of CPCSEA nominee:**

Signature with date

Chairman/Member Secretary of IAEC:

CPCSEA nominee:

(Kindly make sure that minutes of the meeting duly signed by all the participants are maintained by Office)

BIBILIOGRAPHY

1. Medical encyclopedia
2. Times of india ,May23, 2012
3. American medical journal gastroenterol 2002
4. Kumar and clak clinical medicine 5th edition 2002 edited by Parveen kumar, michale clerk
5. http//www.harshulayur.com/chemical-analysis.htm
6. siddha maruthuvam (pothu) 6the edition 2004 by S.K. Kuppusamy mootnaliya
7. IndianJournal of geo-marine sciences vol 41 -Api-2012.
8. Kannusamy Parambarai Vaithiyam 5th edition 2006
9. Gunapadam porutpanpu nool - 2st part thathu vaguppu pg no
10. Gunapadam porutpanpu nool - 1st part mooligai vaguppu pg no 160 7th edition2003
11. United States Of America Department of Agriculture (USDA)
12. Poutiers, J. M. (1998). Gastropods in: FAO Species Identification Guide for Fishery Purposes: The living marine resources of the Western Central Pacific Volume 1. Seaweeds, corals, bivalves and gastropods. Rome, FAO, 1998. page 503.
13. Wikepedia
14. Indian material medica vol 1 A.K. Nadkarunai 2005 vol 2
15. Kulshersthesthestha mayank et ai, International Research Journal Of Pharmacy. ISSN 2230-8407
16. A study on Palagarai Parpam for Anti spasmodic activity and Analgesic and muscle relaxant Properties on suthagavali N.I.S.April,2011.
17. Database on medical plants used in Ayurveda volume, central council for Research in Ayurveda and Siddha.vol 1
18. Kirtikan &BASU Vol IV 2nd edition 2004
19. A critical review on Ayurvethic drug Kapardica (Cypraea moneta linn). D.M.Vasudevan, Sreekumari, Text Book of Biochemistry, Jaypee brother's Medical Publishers(P)ltd,5th edition,313-326.-calcium
20. General Article of Marine Organism inIndian Medicine and their future prospects. Natural Products Radiance Vol 7 (2) 2008 pg 139-145.
21. Harrison's principles of internal medicine.16th edition vol 2 2005 pg no 1808

22. KV Krishna Das,Textbook of Medicine, 5th Edition,Jaypee Brothers Medical Publishers (P) Ltd,2008,984, 989, 990.
23. Dr.Uththamarayan,H.P.I.M.,Thotra Kirama Araichiyum Siddha Maruthuva Varalarum,Department of Indian Medicine and Homeopathy,3rd Edition,2003.pg no 237
24. Noinaadal noi muthal naadal thirathu 1st vol 2003 3rd edition by Dr.Shanmugavelu H.I.M.P pg no 24.
25. Thomas M.Delvin, Text Book of Biochemistry with Clinical correlations,Kaye pace-2011,1086.
26. www.Sciencedaily.com/ release2011/05/110502174231.hmt.
27. www.humihealth into /hugemineral.html
28. www.science daily.com/release/2011/05/110502174231.htm.
29. Clinical Trial in Rheumatology Arthritis and Osteoarthritis , David M.REID, Colin G,MILLER, Krishnan Baburaj pg 281-2008 ISSN-1852338741.
30. Osteoarthrutis by March Miller.
31. Principle of geriatric Phisiotheraphy by Multani pg no 93.
32. Text book of orthopedics 4th edition by John Ebnezer2010
33. T.V.Sambasivam Pillai, Tamil Agarathi, Dept., of Indian Medicine and Homeopathy
34. Pulipani Vaithiyam 500.
35. Sikicharathina deepam pg no 32.
36. Dept., of Indian Medicine and Homeopathy, Saraku Suthi Sei Muraigal,1 edition 2008 pg 32 -Aanaivari ananthan Ph.D .
37. Yakopu Vaithiyam 300,Thamarai Noolagam ,2nd edition,2004

REFERENCES

1. Afaf I, Abuelgasim, Nuha HS, Mohammed AH (2008), Hepatoprotective effect of *Lepidium sativum* against carbontetrachloride induced damage in rats. Res J Animal Veterinary Sci, 3, pp 2023.
2. Aftab A, Pillai KK, Abul KN, Shibli JA, Pal SN, Balani DK. Evaluation of hepatoprotective potential of jigrine post treatment against thioacetamide induced hepatic damage. J Ethnopharmacol 2002;79:35-41.
3. Ajay KG, Neelam M (2006), Hepatoprotective activity of aqueous and ethanolic extract of *Chamomile capitula* in paracetamolintoxicated albino rats. American J Pharmacol Toxicol, 1(1), pp 1720.
4. Anand KK, Gupta VN, Rangari VD, Chandan BK. Structure and hepatoprotective activity of bioflavonoid from Canarium manii. Planta Med 1992;58:493-5.
5. Arulkumaran KS, Rajasekaran A, Ramasamy A, Jegadeesan M, Kavimani S, Somasundaram A (2009), *Cassia roxburghii* seeds protect liver against toxic effects
6. Ashok S. K. , Somayaji S.N. and BairyK.L. Hepatoprotective effects of *Ginkgo biloba* against carbon tetrachloride induced hepatic injury in rats. Indian J Pharmacol 33(2): 260-6 (2001).s
7. Bahar A, Tanveer A, Shah AK. Hepatoprotective activity of Luffa echinata fruits. J Ethnopharmacol 2001;76:187-9.
8. Bardhan P, Sharma SK, Garg NK (1985), in vitro effect of an ayurvedic liver remedy on hepatic enzymes in carbon tetrachloride treated rats. Indian Journal of Medical Research, 82, pp 359364.
9. Bishayee, A., Sarkar, A., Chatterjee, M., . The hepatoprotective activity of Carrot (Daucus carota L) against carbon tetrachloride intoxication in mouse liver. J. Ethnopharmacol. 1995, 47, 69–74.
10. Boyd EH and Bereczky GM. Liver necrosis from paracetamol. Br J Pharmacol. 1966; 26: 606-614.
11. Deb AC. Liver function tests. Fundamentals of biochemistry. 8th edition; Kolkata: 2008:581:6.

12. Gupta AK and Misra N. Hepatoprotective Activity of Aqueous Ethanolic Extract of *Chamomile capitula* in Paracetamol Intoxicated Albino Rats. American Journal of Pharmacology and Toxicology. 2006; 1: 17-20.
13. Gupta, A.K. and N. Misra (2006), Hepatoprotective Activity of Aqueous Ethanolic Extract of Chamomile capitula in paracetamol intoxicated albino rats. A m .J. Pharm. Toxicol., 1, pp 17 20.
14. Harshmohan. The liver, biliary tract, exocrine and pancreas: Textbook of Pathology. 4th ed. New Delhi: Jaypee Brothers Medical Publishers (P) Ltd; 2002:22-4,569-80.
15. Hinson. J.A. Pohl. Monks, Gillete JR. Acetaminophen induced hepatotoxicity. Life Sci 1981; 29: 107-116.
16. HuiMei L, HsienChun T, ChauJong W, JinJin L, ChiaWen L, FenPi C (2008), Hepatoprotective effects of *Solanum nigrum* Linn extract against CCl4iduced oxidative damage in rats. ChemicoBiological Interactions, 171, pp 283–293
17. Irwin. The organization of screening. In: Robert A Turner. Screening methods in Pharmacology. India: Elsevier; 2009. p. 22-40.
18. Jalalpure SS, Patil MB. Hepatoprotective activity of the fruits of *Piper longum*. Indian J Pharm Sci. 2003;65:363.
19. Karan, M., Vasisht, K., Handa, S.S.. Antihepatotoxic activity of Swertia chirata on carbon tetrachloride induced hepatotoxicity in rats. Phytotherapy Research. 1999. 13, 24–30.
20. Krishna V, Mankani KL, Shanthamma C. Evaluation of hepatoprotective activity of stem bark of *Diospyros cordifolia*. Indian J Pharm Sci. 2005;67:106.
21. Kumar G, Sharmila Banu G, Vanitha Pappa P, Sundararajan M, Rajasekara Pandian M. Hepatoprotective activity of Trianthema portulacastrum L. against paracetamol and thioacetamide intoxication in albino rats. J Ethnopharmacol 2004;92:37-40.
22. Kumar SR, Mishra SH. Hepatoprotective activity of the whole plants of *Fumaria indica*. Indian J Pharm Sci. 1997;59:165.
23. Lin, C.C., Yen, M.H., Lo, T.S., Lin, J.M., Evaluation of the hepatoprotective and antioxidant activity of Boehmeria nivea var. nivea and B. nivea var. tenacissima. J. Ethnopharmacol. 1998, 60, 9–17.

24. Marshal M, Kalpan MD. Understanding liver function tests 2007. Obtained from world wide web: http://ocw.tufts.edu/data/48/593572. pdf (cited on12th July 2011).
25. Nkosi CZ, Opoku AR and Terblanche SE. Effect of pumpkin seed (*Cucurbita pepo*) protein isolate on the activity levels of certain plasma enzymes in CCl4-induced liver injury in lowprotein fed rats. Phy.the.Res.2005; 19: 341–345.
26. Prescott, L.F. Wringht. N. Roscoe. P and Brown, S.S. Plasma Paracetomol half-life and hepatic necrosis in patients with paracetomol overdose Lancet. I; 519-522.
27. Rane A, Grampurohit ND. Hepatoprotective activity of *Petrocarpus marsupium* and *Butea frondosa* koen. Indian J Pharm Sci. 1998;65:182.
28. Reitman S and Frankel S. *In vitro* determination of tranaminase activity in serum. Am. J. Clin. Pathol. 1957; 28: 56.
29. Richard OR. A new direction in the study of carbon tetrachloride hepatotoxicity. Life Sci 1983;33:401-8.
30. Roger walker, Cate W. Liver diseases. Clinical pharmacy and therapeutics. Churchil living stone: 4th edtion; London: 2007:221-2.
31. Sanmugopriya, E., Venkataraman,S., Studies on hepatoprotective and antioxidant actions of Strychnos potatorum Linn. seeds on CCl4-induced acute hepatic injury in experimental rats J. Ethnopharmacol., 2006. (105). 154-160.
32. Satyanarayana U, Chalrapani. Liver function tests. Fundamentals of biochemistry Kolkata; 2006:453-8.
33. Seakins A, Robinson DS. The effect of the administration of carbon tetrachloride on the formation of plasma lipoproteins in rats. Biochem J 1963;86:401-7.
34. Sharma A. Chakraborti. K.K., Handa S.S. Anti hepatotoxic activity of some Indian herbal formulation as compared to silymarin Fitoterapia. 1991; 62:299-35.
35. Shashi K, Ramaiah, Apte U, Mehendale HM. Cytochrome P4502E1 induction increases thioacetamide liver injury in diet restricted rats. Drug Meta Dispo 2001;29:1088-95.
36. Suja, S.R., Latha, P.G., Pushpangadan, P., Rajasekharan, S., Evaluation of hepatoprotective effects of Helminthostachys Zeylanica (L.) Hook against carbon tetrachloride induced liver damage in Wistar rats. J. Ethnopharmacol.2004, 92, 61–66.

37. Suresh Kumar SV, Sujatha C, Syamala J, Nagasudha B, Mishra SH. Hepatoprotective activity of extracts from Pergularia daemia Forsk. against Carbon tetrachloride induced toxicity in rats. Phcog Mag 2007;3:11.
38. Tran QI, Adnyana IK, Tezuka Y, Nagaoka T, Tran QK, Kadota S. Triterpene saponins from Vietnamese ginseng (Panax vietnamensis) and their hepatocyte protective activity. J Nat Prod 2001;64:456-61.
39. Vijayan P, Prashanth HC, Dhanraj SA, Badami S, Suresh B. hepatoprotective effect of total alkaloid fraction of Solanum pseudocapsicum leaves. Pharmaceut Biol 2003;41:443-8.
40. Taga MS, Miller EF and Pratt DE. Chia seeds as a source of natural antioxidants. *J. Am. Oil. Chem. Soc*. (1984) 61: 928-931
41. Rajesh L and Mehta Joseph F. Ajowan as a source of natural lipid antioxidants. *Agric. Food Chem*. (1994) 42: 420-1422

ACKNOWLEDGEMENT

I feel immense awe and colossal gratitude in my heart of hearts to God Almighty for making this dissertation have its present form.

I express my sincere thanks to the Vice-Chancellor, The Tamilnadu Dr.MGR medical university Chennai-32.

I express my sincere thanks to Prof. **Dr.K.Manickavasagam M.D(S),** Director National Institute of Siddha, Tambaram sanatorium,Chennai-47.

I express my gratitude to **Dr.M.Rajasekaran M.D.(S).,** Associate Prof., Head of the Department of Gunapadam i/c, National Institute of Siddha, Tambaram sanatorium, Chennai-47, for his encouragement, suggestions and valuable guidance in this dissertation work.

I express my sincere thanks to **Dr. A.Rajendra kumar M.D.(S).,** Associate professor, Gunapadam department, NIS,Chennai-47, for his suggestions.

I express my sincere thanks to **Dr .S.Visweswaran M.D.(S).,** Lecturer, Gunapadam department, NIS,Chennai-47, for his suggestions.

I express my sincere thanks to **Dr.S.Sivakumar M.D.(S).,** Lecturer, Gunapadam department, NIS,Chennai-47, for his suggestions.

I express my sincere thanks to **Dr.A.Mariappan M.D.(S).,** Lecturer, Gunapadam department, NIS,Chennai-47, for his suggestions.

I express my grateful thanks to **Dr.J.Anbu Ph.D.,**Vels college of pharmacy, Pallavaram, Chennai.

I express my sincere thanks to **Dr.D.Aravind M.D.(S) M.Sc.,** Assistant Professor, Medicinal Botany,NIS,Chennai-47.

I express my sincere thanks to **Dr.V.Suba M.Pharm, P.hD.,** Assistant Professor in Pharmacology, NIS, Chennai-47.

I express my sincere thanks to **Dr.Muthuvel, Assistant Professor,** Biochemistry Dept.,NIS,Chennai-47.

I express my sincere thanks to **Mr.M.Subramanian MSc,**SRO,NIS,Chennai-47, for his guidance in statistical analysis.

I express my grateful thanks to **Dr.C.Saravana babu, M.Pharm.,Ph.D.,** SRMC, Porur, Chennai.